DU® Series 700 User's Guide

A24014-AA

August 2005

Beckman Coulter, Inc.
4300 N. Harbor Blvd., Fullerton, CA 92834-3100
© Copyright 2005 Beckman Coulter, Inc.

Copyright, Licenses and Trademarks

Copyright

© Beckman Coulter, Inc., 2005. All rights reserved. No part of this publication may be reproduced, transcribed, transmitted, or translated into any language in any form by any means without the written permission of Beckman Coulter, Inc.

Licenses and Trademarks

Beckman Coulter® and DU® are registered trademarks of Beckman Coulter, Inc.

Microsoft and Windows are registered trademarks of Microsoft Corporation in the United States and other countries.

Table of Contents

Section 1 Introduction

General Information 1
- Proper Use 1
- About this Manual 1
- Product Compliance 1

Safety Precautions 2
- Electrical Safety 2
- Safety Against Risk of Fire 2
- Chemical and Biological Safety 3
- Source Lamp Safety 3
- Use of Hazard Information 3
- Precautionary Labels 3
- Waste Management and Recycling 4

Section 2 System Description

Systems and Functions 5

Design 6
- Front and Back View 6
- Display 6
  - Tips for Using the Touch Screen 7
- Ports and Power 8
- Beam Path 9
- Source Compartment 10

Unpacking the Instrument 10

Operating Environment 10

Power Up / Applying Power 11

Section 3 Installation

Cell Compartment 13

Cell Holders 13
- Standard Cell Holder 13
  - Standard Cell Holder Installation 14
- 50 µL Single Cell Holder 15
  - 50 µL Single Cell Holder Installation 15
- Turbidity Cell Holder 16
  - Turbidity Cell Holder Installation 16
- Multicell Holder 17
  - Multicell Holder Installation 17
- Carousel Holder (Sample Changer) 18

Carousel Holder Installation 18
Modules 19
Sipper Module 19
Sipper Module Installation 21
Peltier Temperature Control Module 25
Peltier Temperature Control Module Installation 25
Switching the Power On 27
Language Selection 27
Self-Check 27
Main Menu 28
Alphanumeric Keypad 30
Instrument Setup Mode 31
Operator ID 31
Sample ID 32
Date and Time 33
Display and Sound Preferences 34
Lamp Control 34
USB Port 36
Password 38
Carousel and Module Options 40
Carousel 40
Sipper Module 41
Peltier Module (Temperature Control) 42

Section 4 User Programs
Storing a User Program 43
Recalling a User Program 44
Deleting a Stored User Program 44

Section 5 Fixed Wavelength Mode
Parameter Setup 45

Section 6 Wavelength Scan Mode
Parameter Setup 51
Scanning 55

Section 7 Kinetic/Time Mode
Parameter Setup 57
Taking the Kinetics Readings 58
Analysis of a Kinetic Run 59

Section 8 Single Component Analysis Mode (SCA)

Parameter Setup 61
Standard Curve Setup 61
Coefficients Setup 64
Reading Standards 65
Analyzing Samples 67

Section 9 Protein Assay Analysis

Method Description 69
Bradford 69
Lowry (High) 69
Lowry (Low) 70
Biuret 70
UV280 70
Colloidal Gold 70
Bicinchoninate (BCA) 70
Parameter Setup 71
Standard Curve Setup 71
Coefficients Setup 74
Reading Standards 75
Analyzing Samples 76

Section 10 Nucleic Acid Analysis

Method Description 79
260/280 Ratio 79
Single Ratio 79
Double Ratio & Concentration 79
dsDNA, ssDNA, and RNA 79
Oligo DNA Long and Oligo RNA Long 80
Oligo DNA Short and Oligo RNA Short 80
Parameter Setup 81
260/280 Ratio and Single Ratio 82
Double Ratio & Concentration 83
dsDNA, ssDNA, and RNA 85
Oligo DNA Long and Oligo RNA Long 86
Oligo DNA Short and Oligo RNA Short 87
Analysis of Samples 88
Calculations 91
Background Correction and Net Absorbance 91
Concentration Calculations 91
Molecular Weight (Oligo DNA Long/Short) 92

Molecular Weight (Oligo RNA Long/Short) 92
Molar Extinction Coefficient (Oligo DNA Long) 93
Molar Extinction Coefficient (Oligo DNA Short) 93
Molar Extinction Coefficient (Oligo RNA Long) 94
Molar Extinction Coefficient (Oligo RNA Short) 94
Extinction Coefficient (Oligo DNA/RNA Long/Short) 95
Concentration (Oligo DNA/RNA Long/Short) 95
Melting Temperature (Oligo DNA Long) 96
Melting Temperature (Oligo DNA Short) 96

Section 11 % Dye Incorporation

Method Description 99
Parameter Setup 100
Analysis of Samples 101

Section 12 DNA/Protein Tools Mode

Calculations 103
Protein Concentration from Amino Acid Sequence Data 104
Radioactive Decay Correction 106
Conversions 108
Micrograms or dsDNA to Picomoles 108
Example for Conversion Functions 109
Picomoles of dsDNA to Micrograms and Nanograms 109
Micrograms/mL of Oligonucleotides (ssDNA) to Picomoles/µL 110
Picomoles/µL of Oligonucleotides (ssDNA) to Micrograms/mL 110
Micrograms of Linear DNA to Picomoles of Ends 111
Molar Ratio of Insets to Vector for Ligations 111
Nucleic Acid Concentration from Abs@260 112
Protein Molar Conversion 113
Protein Coding Capacity 114
Temperature Conversion 115
Tables 115
Nucleotide Naming Conventions 115
Standard Genetic Code 116
Amino Acid Codes and Masses 116
DNA/Protein Sizes 116
DNA Sequence Triplets (Decoder) 116
Solubility, Density, and pI of Amino Acids 116
Hydrophobicity of Amino Acids 116
Radioactivity Units 116
Metric Prefixes 116

Section 13 Carousel, Sipper, and Peltier Operation

Carousel Holder 117
Fixed Wavelengths Measurements 117
Wavelength Scan 118
Kinetics/Time 119
Sipper Module 120
Fixed Wavelengths Measurements 120
Wavelength Scan 121
Kinetics/Time 122
Peltier Module 123
All Modes 123

Section 14 Store, Recall, Send, and Delete Data

Storing Data 125
Storing Data - Auto/Manual 125
Single Reading Mode 125
Continuous Reading Mode 126
Storing Scans (Wavelength Scan) 126
Storing Kinetic Data (Kinetics/Time) 127
Recalling Data, Scans, and Kinetic Runs 127
Recalling Data 127
Recalling Scans (Wavelength Scan) 128
Recalling Kinetic Data (Kinetics/Time) 130
Sending Data, Scans, and Kinetic Runs 131
Sending Data 131
Sending Scan Data (Wavelength Scan) 132
Sending Kinetic Data (Kinetics/Time) 133
Deleting Data, Scans, and Kinetic Runs 133
Deleting Data 133
Deleting Scan Data (Wavelength Scan) 135
Deleting Kinetic Data (Kinetics/Time) 135

Section 15 Maintenance

Cleaning Requirements 137
Spectrophotometer 137
Display 137
Cell Cleaning Instructions 137
Cell Description 137
Cell Cleaning Instructions 138
Cell Deterioration 139
Flowcell Maintenance/Cleaning Instructions 139
Flowcell Cleaning Instructions 139

Conditioning a new Flowcell 140
Cleaning Procedure - Daily 140
Cleaning Procedure - Contaminated Flowcells 140
Cleaning Procedure - Severely Contaminated Flowcells 141
Other information 141
Changing Lamps 142
Changing the Tungsten Lamp 142
Changing the deuterium lamp (UV) 143
Changing the Fuse 144
Changing the Filter Pad 145

Section 16 Troubleshooting
Power Up Diagnostics 147
Error Messages 148
Other Error Conditions 149

Section 17 System Check
Call System Check from the Main Menu 151
Instrument Information 151
Upgrading the Instrument Software 152
Optical Checks 152
Wavelength Check 153
Noise Check 154
Stray Light Check 154
Absorbance Check 155
Drift Check 156
Lamp History 157
Factory Service 157

Section 18 Specifications
Performance Specifications 159
Physical and Environmental Specifications 159
Power and Other Specifications 160

Section 19 Replacement Parts, Service Centers, and Warranty
Replacement Parts, Supplies, and Accessories 161
Repair Service/Beckman Coulter Sales and Service Offices 163
Warranty 163

Introduction

1.1 General Information

The spectrophotometer has been designed in accordance with the current state of the technology and the acknowledged safety regulations.

The manufacturer certifies that this instrument was thoroughly tested, inspected, and found to meet its published specifications when it was shipped from the factory.

Nevertheless, under certain circumstances users may be at risk, or the proper functioning of the instrument may be impaired.

WARNING If risk-free operation is not possible, the instrument should not be switched on or it should be properly switched off and secured against being switched on unintentionally.

This is the case when:

- The instrument is visibly damaged.
- An electrical failure occurs.
- The temperature is above 70°C.
- Transport damage occurs.

Proper Use

The spectrophotometer is solely intended for carrying out individual measurements or series of measurements for laboratory analysis.

Beckman Coulter, Inc. cannot accept liability for any harm caused to persons or property as a consequence of using the instrument for any purpose other than those defined in the manual.

About this Manual

This manual contains all the information needed to enable the instrument to be operated without any problems throughout its life span.

The manual is intended for use by the personnel who operate the instrument. Accident- and trouble-free operation of the instrument can only be assured by adhering strictly to the instructions in the manual. Such adherence results in less down-time and lower repair costs, and increases the lifesaving of the instrument.

Product Compliance

This equipment complies with Class A limits for the FCC and EN 55011 requirements.

1.2 Safety Precautions

Please read this entire manual before unpacking, setting up, or operating this instrument. Pay particular attention to all danger and caution statements. Failure to do so could result in serious injury to the operator or damage to the equipment.

Do not use or install this equipment in any manner other than what is specified in this manual.

Besides following the instructions in this manual, users must comply with the general safety and accident prevention regulations of the country in which the instrument is used.

Electrical Safety

To reduce risks of electric shock, this equipment is equipped with a three-wire electrical cord and plug to connect the equipment to earth ground. To preserve this safety feature:

- Make sure the matching wall outlet receptacle is properly wired and earth grounded.
- Never use a three- or two-wire isolating plug adapter.
- Never use a two-wire extension cord or a non-grounding type multiple outlet receptacle strip.
- Any servicing of this equipment that requires removing covers or panels can expose parts that may cause electric shock or personal injury. Refer such servicing to qualified service personnel.

Safety Against Risk of Fire

- Certain electrical circuits within this equipment are protected by fuses against over-current conditions. For continued protection against a risk of fire, replace fuses only with the same type and rating specified.

Chemical and Biological Safety

Normal operation of this instrument may involve using toxic, flammable, or biologically harmful chemicals. Users must take these precautions:

- Observe all cautionary information printed on the original solution containers prior to using them.
- Handle infectious samples in accordance with good laboratory procedures and methods to prevent spread of disease.
- Dispose of all waste solutions in a proper manner.
- When using hydrochloric acid to clean the flow cell, observe standard laboratory safety procedures. Wear protective eye covering and immediately wash any splashes from skin or clothing with plenty of water.
- Since concentrated Trace-Klean is a highly alkaline solution, handle it with care.

Source Lamp Safety

The source lamps operate at high temperatures.

- To avoid a possible electric shock, disconnect the instrument from the power source before servicing the lamps.
- To prevent a possible burn, allow the lamp(s) to cool at least 30 minutes before handling.
- The UV lamp generates UV light. Do not look directly at an operating lamp without wearing UV protective eye glasses.

Use of Hazard Information

The following symbols appear in the manual:

WARNING Users must comply with this warning for their own safety. Failure to do so may result in an injury.

WARNING This symbol, if noted on the instrument, indicates a hot surface.

NOTE A note contains important information which users must take into account when handling the instrument.

Precautionary Labels

Read all labels and tags attached to the instrument.

Personal injury or damage to the instrument could occur if not observed.

This symbol, if noted on the instrument, refers to the instruction manual for operational and/or safety information.

Waste Management and Recycling

A016608L.EPS

This symbol is required in accordance with the Waste Electrical and Electronic Equipment (WEEE) Directive of the European Union. The presence of this marking on the product indicates that the device:

1. was put on the European Market after August 13, 2005 and
2. must not be discarded using the municipal waste collection system of any member state of the European Union.

Customers must understand and follow all laws regarding the proper decontamination and safe disposal of electrical equipment. Please contact your dealer or local Beckman Coulter office for details on the take-back program that will facilitate the proper collection, treatment, recovery, recycling, and safe disposal of Beckman Coulter products bearing this label.

System Description

2.1 Systems and Functions

The DU Series 700 Spectrophotometer is a complete scanning UV/Visible spectrophotometer with a bandwidth of 3 nm and a wavelength range of 190 to 1100 nm. The instrument comes with a complete set of application programs and multi-language support.

The DU Series 700 includes two models:

- DU 720 General Purpose
- DU 730 Life Science

The DU 720 contain the following modes:

- Fixed Wavelength
- Wavelength Scanning
- Time-based Kinetic
- Single Component Analysis

The DU 730 contains the following modes:

- Fixed Wavelength
- Wavelength Scanning
- Time-based Kinetic
- Single Component Analysis
- Protein Assay Analysis
- Nucleic Acid Analysis
- % Dye Incorporation
- DNA/Protein Tools

The system comes with a Standard Single Cell Holder for a 1-cm rectangular cuvette. A test tube holder is fixed in the sample compartment. Other sampling devices, such as a 7-Cell Carousel, a Sipper Module, and a Peltier Temperature Control Module are optional.

You can use the DU Series 700 for testing samples in the visible and ultraviolet wavelength spectrum. A halogen gas-filled tungsten lamp produces light in the visible spectrum (320 to 1100 nm), and a deuterium lamp produces light in the ultraviolet spectrum (190 to 360 nm).

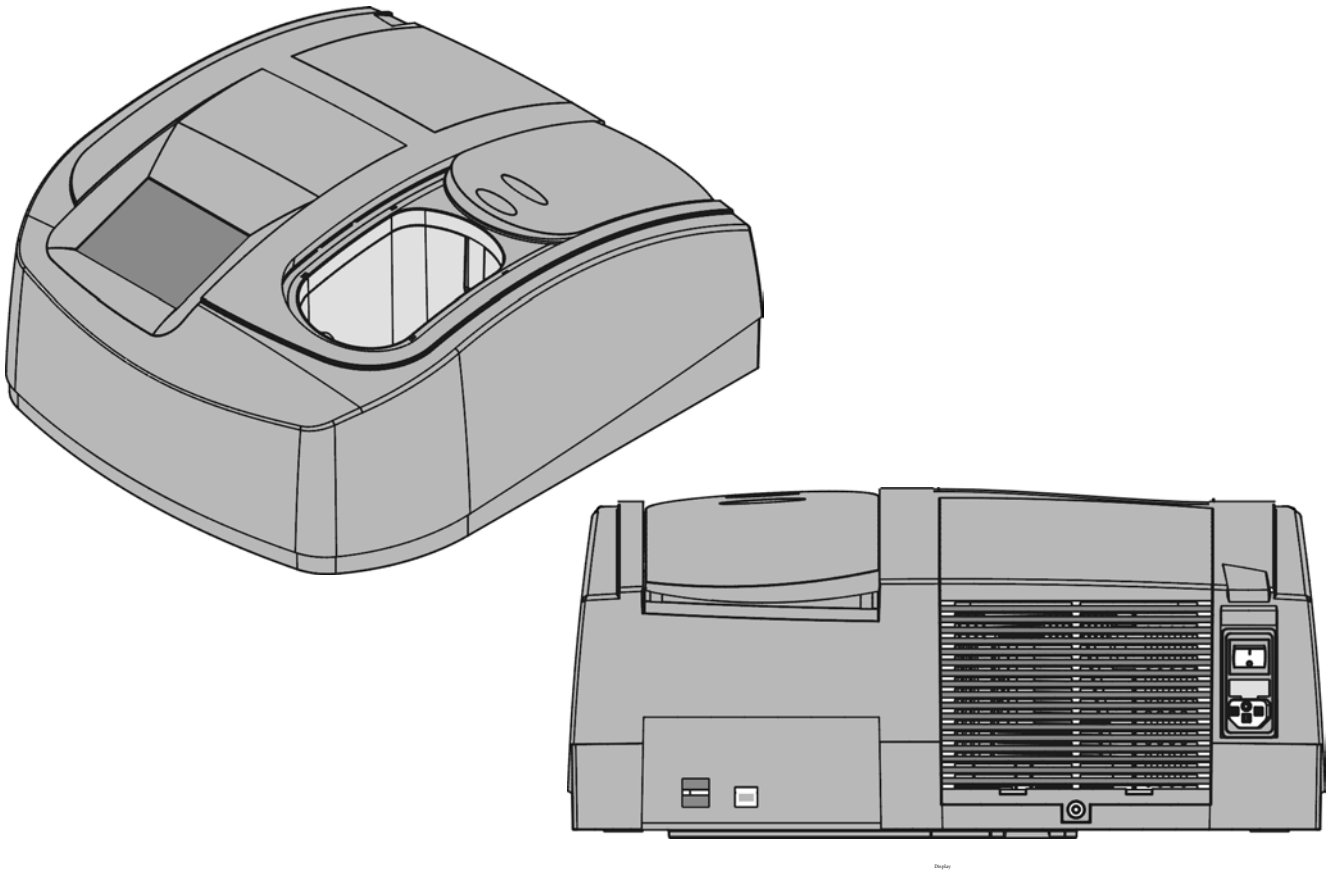
The DU Series 700 Spectrophotometer provides digital readouts in Absorbance or %Transmittance as well as calculated units, depending on the mode. USB ports for printer, keyboard, and memory devices are standard.

When you select a user-generated or programmed method, the on-screen menus and prompts direct you through the test. You can also use these menus to generate reports, run statistical evaluations of generated calibration curves, and report instrument diagnostic checks.

2.2 Design

Front and Back View

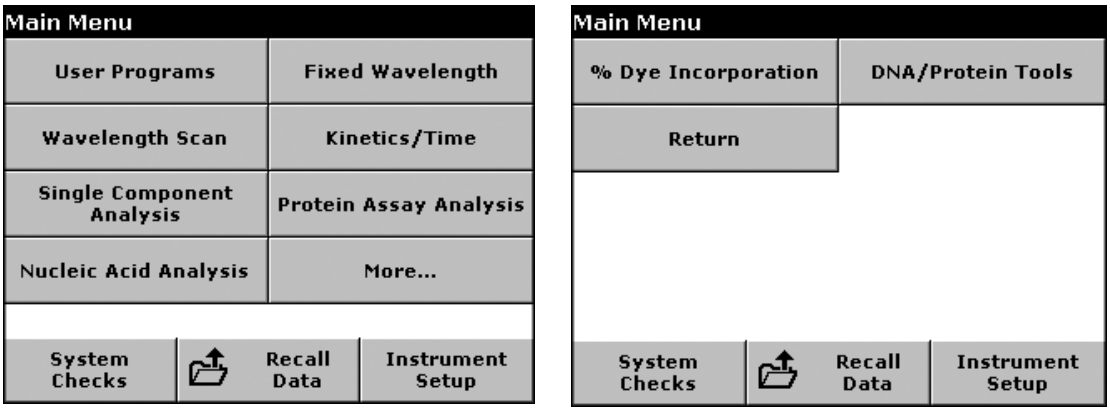
Figure 2.1 Front and Back View



Display

The display is a touch-screen device and the content changes as you select different modes of operation. Figure 2.2 shows the two main menu screens with the available application programs of the DU 730.

Figure 2.2 Main Menu Screens



Tips for Using the Touch Screen

The entire screen is touch-activated (touch screen). Spend a few minutes touching various items on the screen to see how they work.

- Take time to touch the individual fields lightly to get familiar with their functions.
- To make a selection, touch the screen with your fingernail, fingertip, pencil eraser, or a stylus.

NOTE Only the supplied stylus ensures efficient use of the touch screen!

WARNING Do not touch the screen with a sharp object, such as the tip of a ball point pen!

WARNING Do not place anything on top of the cover; doing so could scratch it!

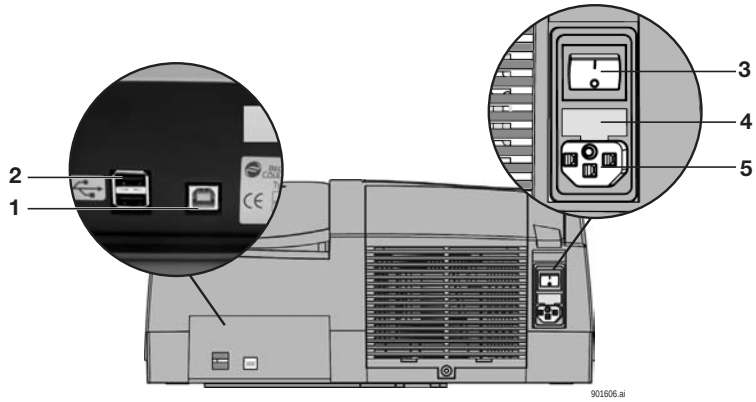
- Touch keys, words, or icons to select them.
- Use scroll bars to move up and down long lists very quickly. Touch and hold the scroll bar, then move your fingertip up or down to move through the list.
- Highlight an item from a list by touching it once. After you select the item, the screen displays the selected item in reversed text (light text on a dark background).

Ports and Power

The power connector and power switch are locate on the back wall of the instrument.

The DU Series 700 Spectrophotometer has three USB ports as a standard feature. They are located on the back wall of the instrument. These USB ports enable you to output data and graphics to a printer or memory device.

Figure 2.3 Ports and Power

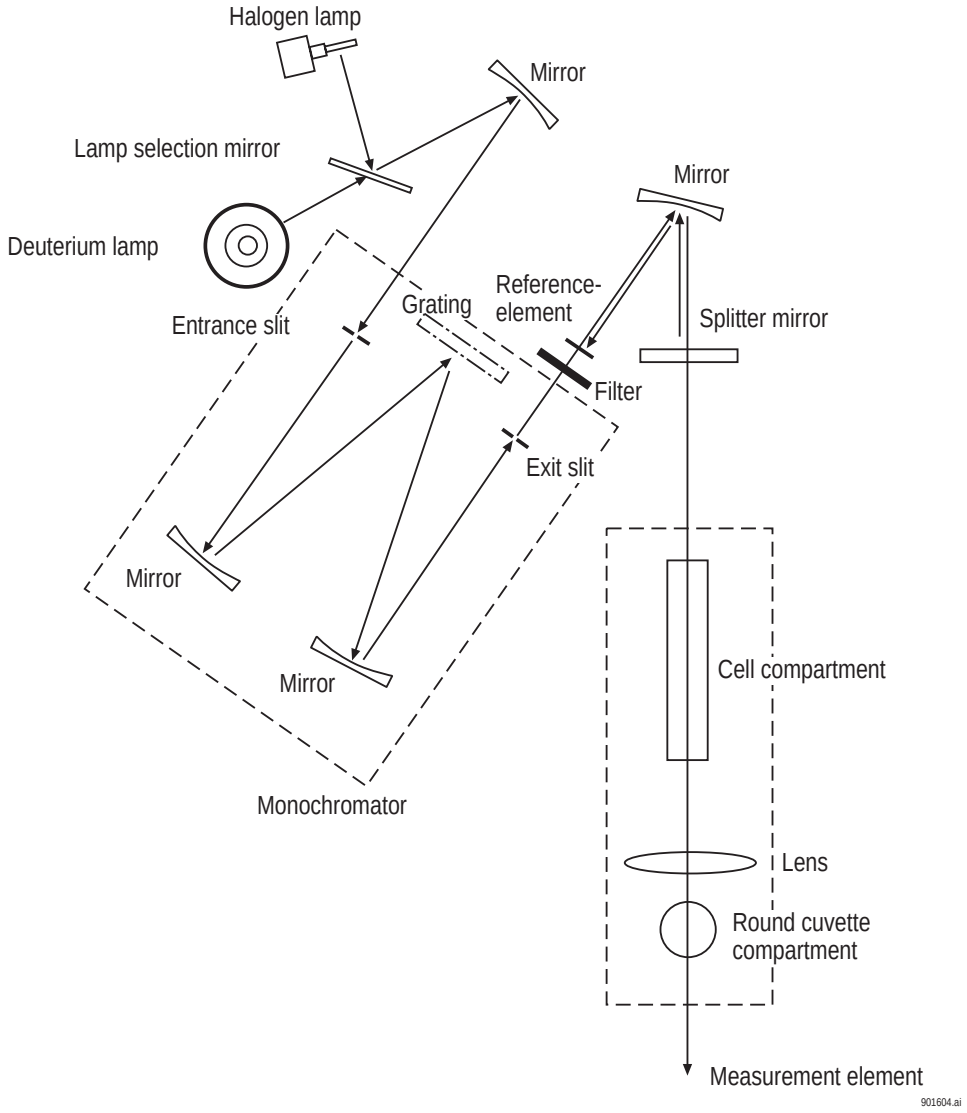


1	Downstream ("B") USB connector
2	Two upstream ("A") USB connectors for Printer, Keyboard, and/or Memory Device.
3	On/Off switch
4	Fuse
5	Plug-in Power Supply

Beam Path

The following schematic diagram shows the beam path of the Spectrophotometer.

Figure 2.4 Beam Path



Source Compartment

The source compartment is on the left side behind the display inside of the instrument. It is provided with ventilation on the back side.

The lamp compartment contains the halogen and deuterium (UV) lamps.

A fan used for cooling the sample compartment and electric components is mounted on the back side. The ventilation system operates automatically.

In general, you should maintain a clear space of at least 15 cm (6 inches) around the instrument for safety. Good ventilation prevents the electronic components from overheating, which helps to extend the life span of the instrument.

WARNING The cover can become hot, especially when a deuterium lamp is used! Do not place anything on top of the cover!

For information on replacing halogen and deuterium lamp, see "Changing Lamps" on page 142.

2.3 Unpacking the Instrument

1. Remove the DU Series 700 Spectrophotometer and accessories from the shipping container.
2. Inspect each item for any damage that might have occurred during shipment.
3. Verify that all items listed in Standard Accessories are included. If any items are missing or damaged, contact your regional Beckman Coulter office or distributor.
4. Do not send the instrument back without previous arrangement.
5. Place the instrument firmly on an even table surface.

2.4 Operating Environment

The following conditions are necessary to ensure that the instrument runs smoothly and has a long life.

- Maintain an ambient temperature of 10 to 40ºC (50 to 104ºF) for proper instrument operation.
- Maintain the relative humidity at less than 90%; moisture should not condense on the instrument.
- Leave at least a 15 cm (6 inch) clearance at the top and on all sides for air circulation to avoid overheating electrical parts.

NOTE Protect the instrument from temperature extremes, including heaters, direct sunlight, and other heat sources.

2.5 Power Up / Applying Power

- Plug the supplied power cord into the connector on the back of the instrument and a main socket
  (100 – 120 V~ / 200 - 240 V~ / 50 – 60 Hz).

NOTE The unit is shipped with a UL/CSA approved 115 Vac power cord with a NEMA 5-15P style plug (North America) or a 230 Vac harmonized power code with a continental European plug.

- Use only earthed sockets.
- Check the power cable for damage before use.

Installation

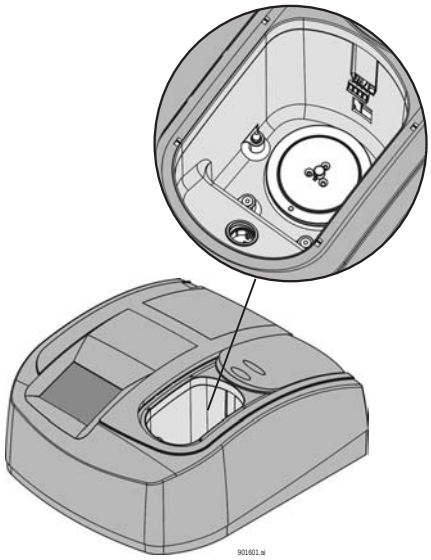
3.1 Cell Compartment

The spectrophotometer has been designed in accordance with the current state of the technology and offers a choice of different modules and interchangeable cell holders for specific applications.

To prevent external light from interfering with the measurement, the cell compartment has a light-proof cover.

NOTE Close the cover before taking a measurement!

Figure 3.1 Cell Compartment

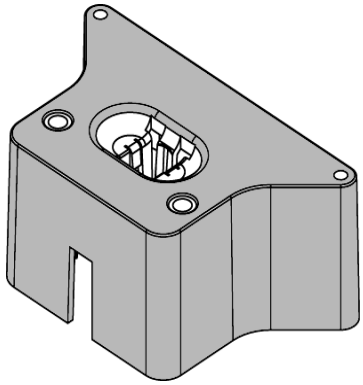


3.2 Cell Holders

Standard Cell Holder

The DU Series 700 Spectrophotometer comes equipped with a Single Cell Holder, which is the Standard Holder. It holds one standard 1 cm rectangular cuvette.

Figure 3.2 Standard Cell Holder

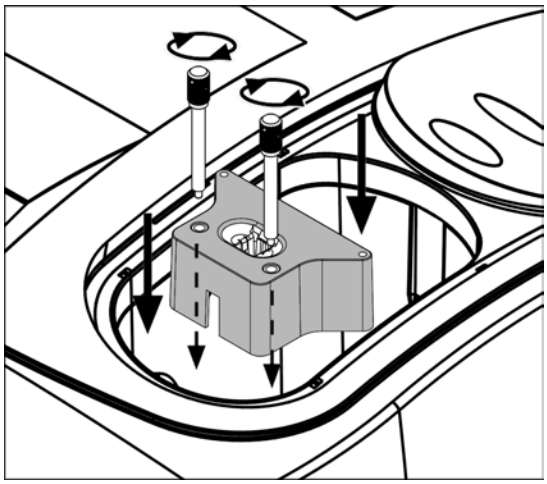


Standard Cell Holder Installation

To assemble the Standard Cell Holder:

1. Open the cell compartment.
2. Insert the cell holder into the cell compartment (see Figure 3.3.)
3. Take care to position the holder exactly. The metal pins of the cell compartment should lock into the holder.
4. Secure the front part of the holder with the two locking screws.

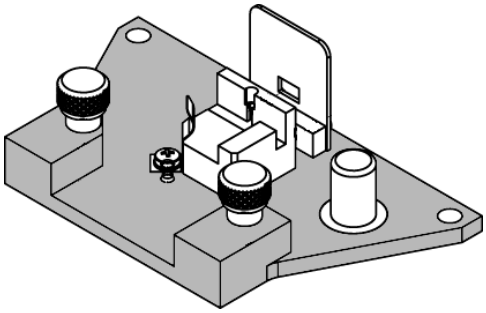
Figure 3.3 Installing the Standard Cell Holder



50 µL Single Cell Holder

The 50 µL Single Cell Holder only accepts 50 µL Microcells from Beckman Coulter, Inc.

Figure 3.4 50 µL Single Cell Holder

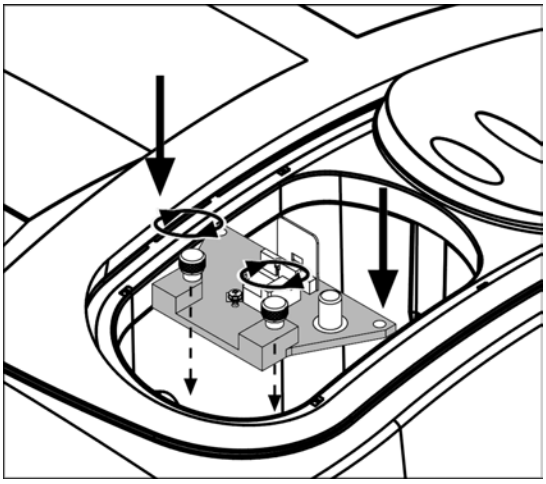


50 µL Single Cell Holder Installation

To assemble the 50 µL Single Cell Holder:

1. Open the cell compartment.
2. Insert the cell holder into the cell compartment in such a way that the locking screws on the holder are at the front.
3. Take care to position the holder exactly. The metal pins of the cell compartment should lock into the holder.
4. Secure the front part of the holder with the two locking screws.

Figure 3.5 Installing the 50 µL Single Cell Holder

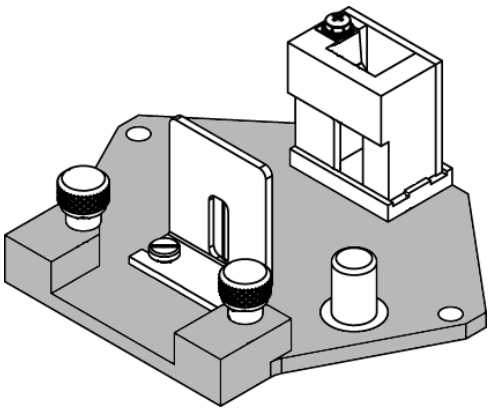


Turbidity Cell Holder

The Turbidity Cell Holder is designed to give accurate quantitative measurements of light scatter in the DU Series 700 Spectrophotometer with conventional forward-optics design.

Use this cell holder when you want to quantitate bacterial cultures or other turbid samples by absorbance measurements.

Figure 3.6 Turbidity Cell Holder

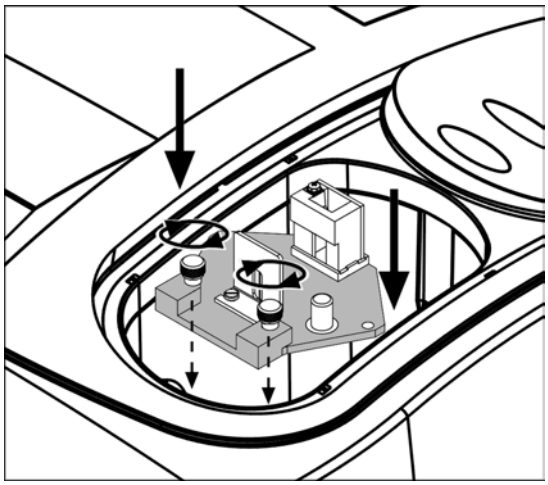


Turbidity Cell Holder Installation

To assemble the Turbidity Cell Holder:

1. Open the cell compartment.
2. Insert the cell holder into the cell compartment in such a way that the locking screws on the holder are at the front.
3. Take care to position the holder exactly. The metal pins of the cell compartment should lock into the holder.
4. Secure the front part of the holder with the two locking screws.

Figure 3.7 Installing the Turbidity Cell Holder



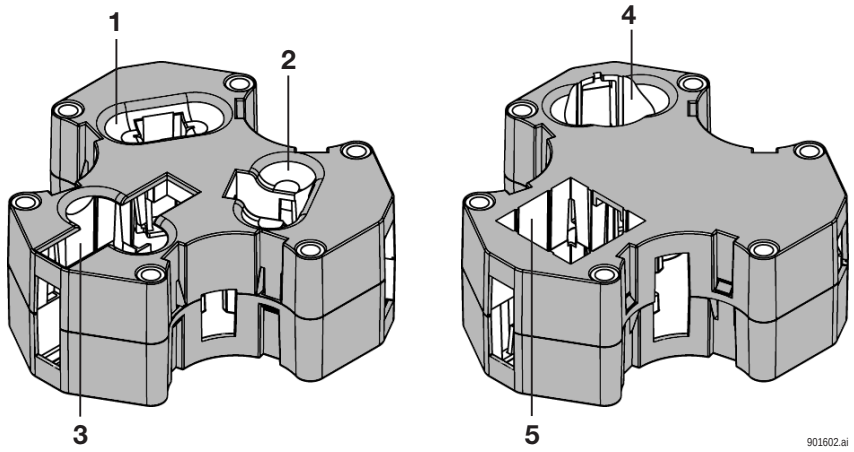
Multicell Holder

The Multicell Holder can accommodate the following cuvette types:

- 10, 20, and 50 mm rectangular cuvettes
- 1-inch round cuvettes
- 1-inch rectangular cuvettes

You can use only one cuvette type for a measurement. On the top and bottom of the Multicell Holder are a variety of openings that accommodate different types of cuvettes. Beside each opening is printed the type of cuvette for which it is intended.

Figure 3.8 Multicell Holder (Top and Bottom)



1	10 mm rectangular cuvette
2	20 mm rectangular cuvette
3	50 mm rectangular cuvette
4	1-inch round cuvette
5	1-inch rectangular cuvette

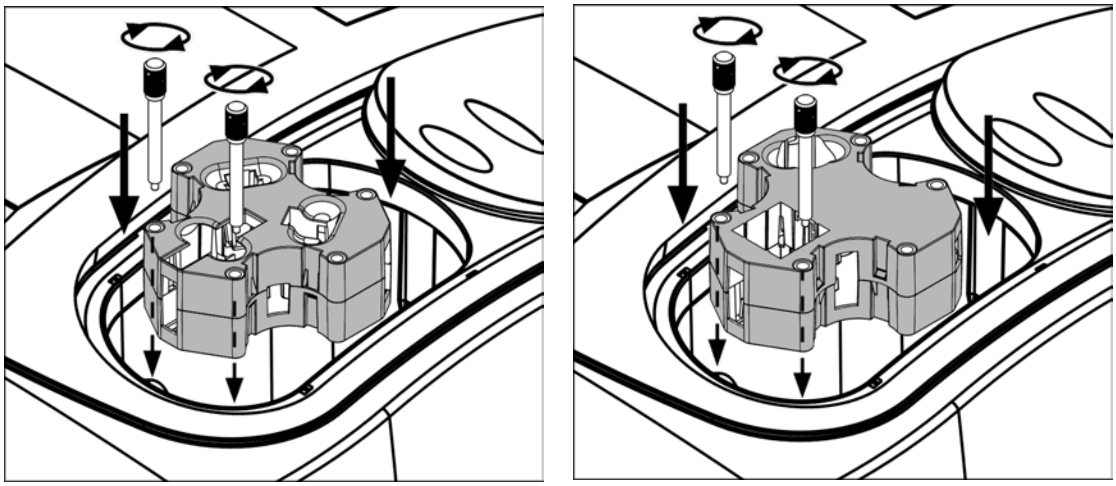
Multicell Holder Installation

To assemble the Multicell Holder:

1. Open the cell compartment.
2. Identify the correct opening for the selected cuvette type in the Multicell Holder.
3. Insert the Multicell Holder in the cell compartment with the cell opening at the front so you can read the name of the selected cuvette type directly.

4. Secure the holder with two locking screws.

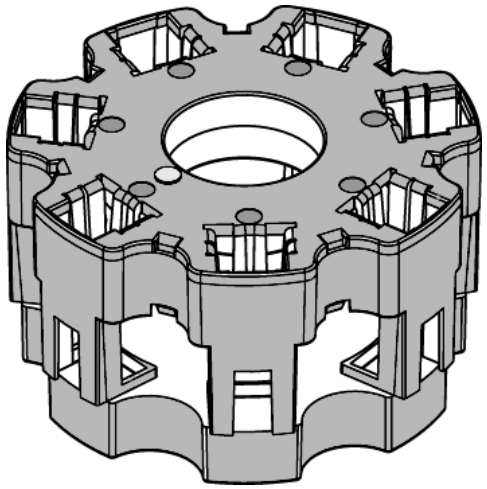
Figure 3.9 Installing the Multicell Holder



Carousel Holder (Sample Changer)

The Carousel Holder allows you to load up to seven cuvettes of solution into the instrument for analysis at one time. The cuvettes can be various combinations of blanks and samples. Use the Setup Mode to activate the Carousel Options and set up the number of cell positions used and the orientation of blanks and samples.

Figure 3.10 Carousel Holder



Carousel Holder Installation

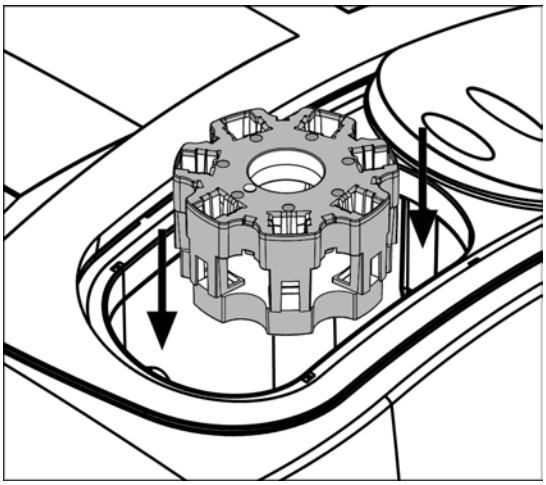
To insert the Carousel Holder:

1. Open the cell compartment.
2. Place the Carousel Holder on the rotatable attachment on the bottom of the cell compartment so that the marking faces upward.

3. Take care to position the holder exactly. The markings on the holder and the rotatable attachment must line up exactly.

4. Turn the holder slightly to the left or right until the guide key locks into position. This establishes contact with the instrument. (For instrument setup options and procedures, see "Carousel and Module Options" on page 40).

Figure 3.11 Installing the Carousel Holder



3.3 Modules

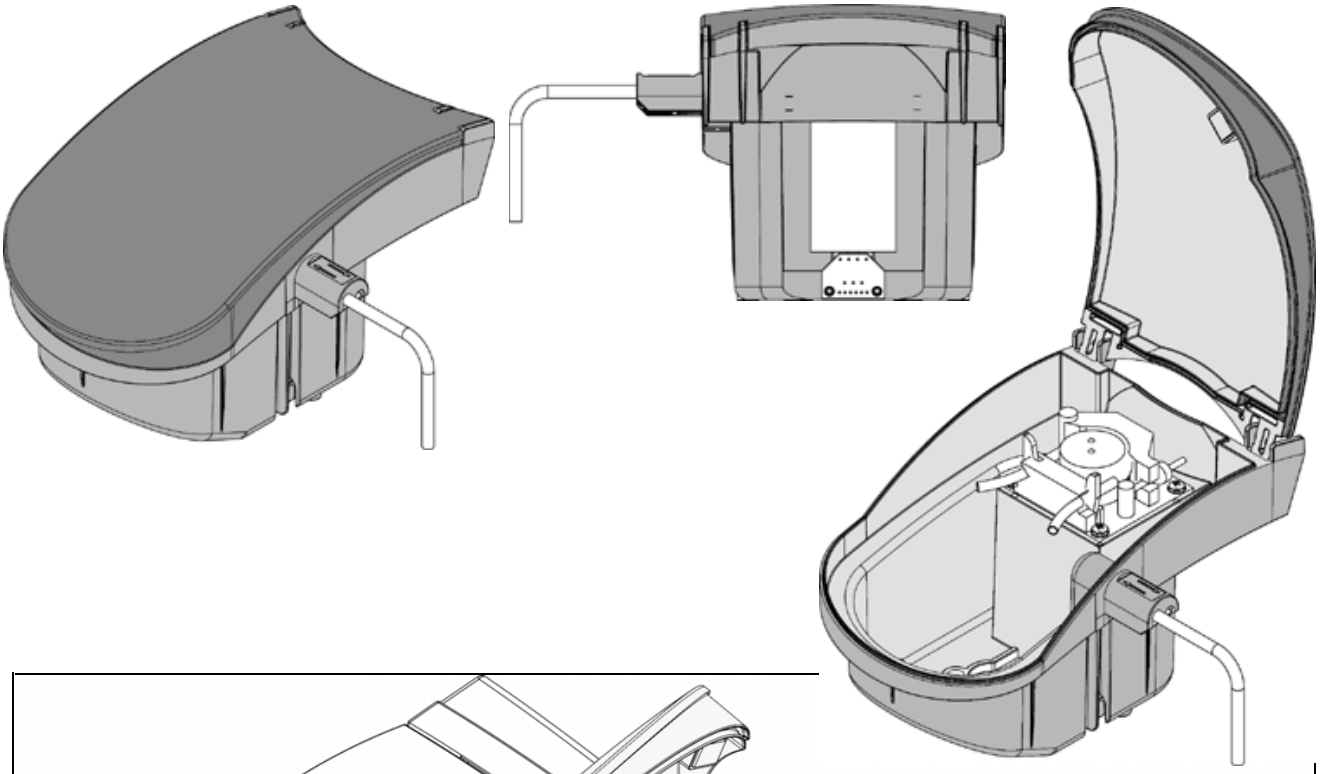
This section describes how to install the Sipper and Peltier Temperature Control Modules.

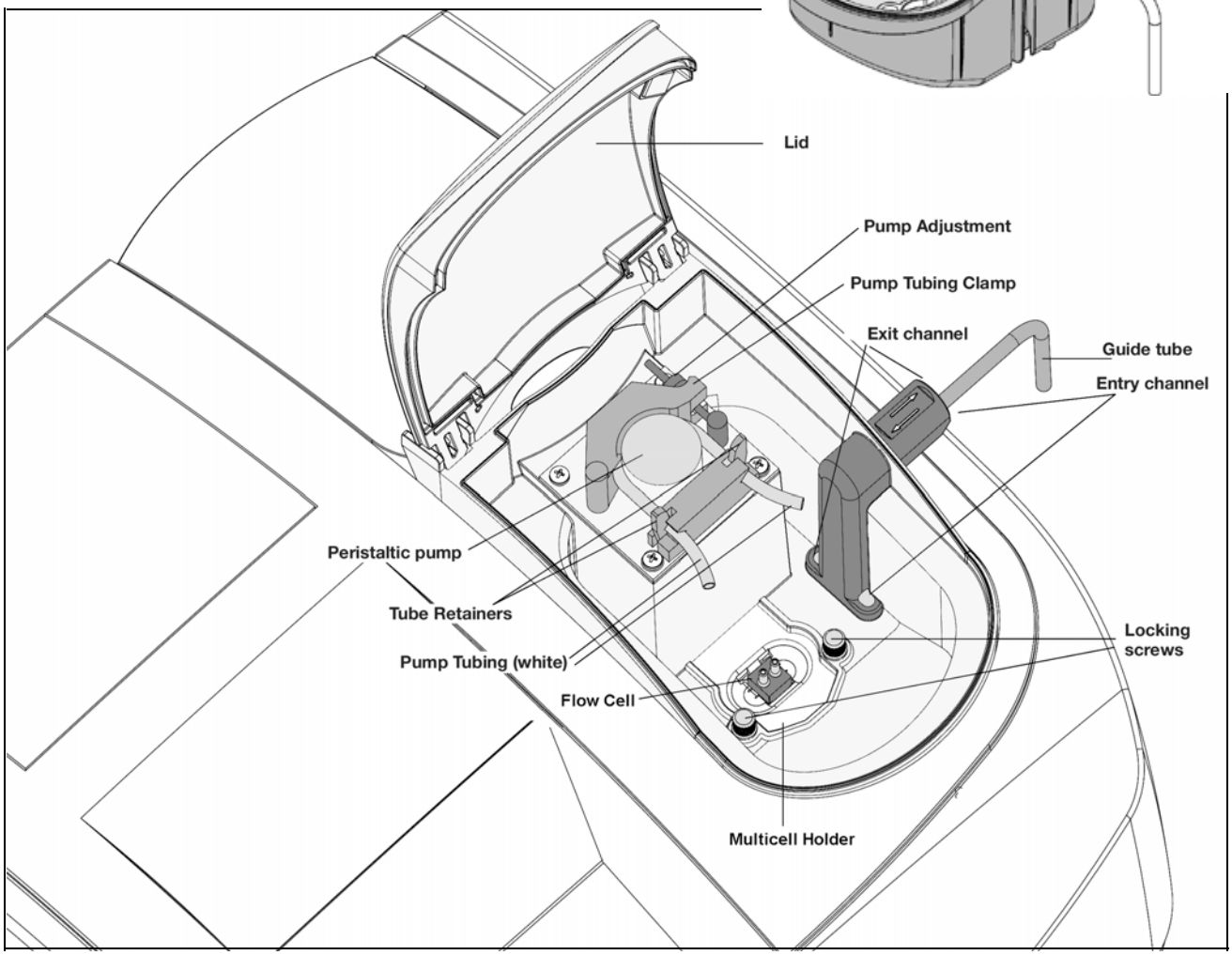
Sipper Module

The Sipper Sampling Module uses a peristaltic pump to aspirate samples into a flow cell for readings. After the module takes a reading, it returns the sample or dumps it to waste.

The Sipper Sampling Module provides improved measurement accuracy, because the same optical characteristics exist for both blanking and reading, and when comparing measurements of different samples. The module eliminates errors that might occur from optical differences between individual vials because it takes every reading in the same vial.

Figure 3.12 Sipper Module



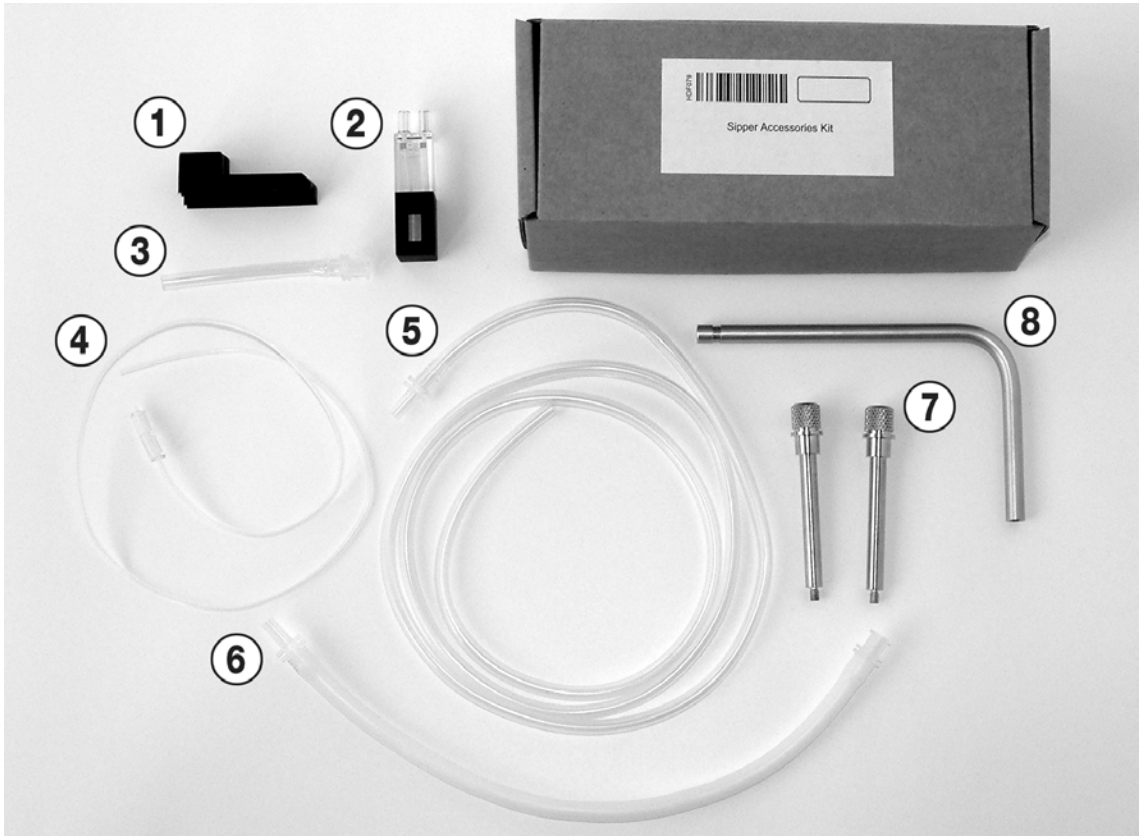


Sipper Module Installation

Remove the Sipper Module from the shipping container and inspect for any damage that may have occurred during shipment. All models are shipped with the following:

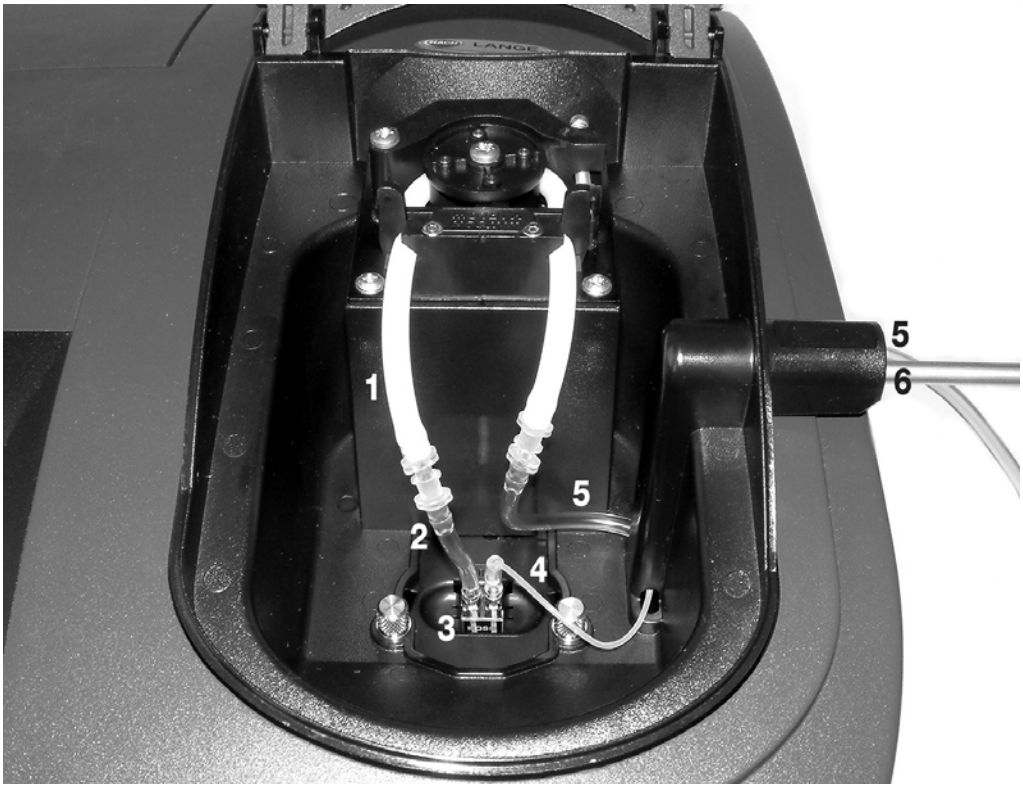
- Sipper Module
- Sipper Accessories Kit (see Figure 3.13)

Figure 3.13 Sipper Accessories Kit



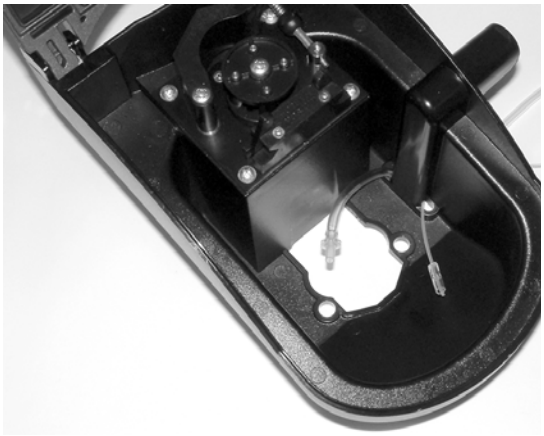
1	Rubber fitting
2	Flow Cell
3	Outlet Connector (from pump to drain)
4	Sample/Inlet Tubing
5	Drain/Waste Tubing
6	Pump Tubing (white)
7	2 locking screws
8	Guide tube

Figure 3.14 Sipper Module with Tubes in Place



1	Pump Tubing (white)
2	Outlet Connector (from pump to drain)
3	Flow Cell
4	Sample/Inlet Tubing
5	Drain/Waste Tubing
6	Guide tube

Before you install the Sipper Module in the DU Series 700 Spectrophotometer, you need to connect a number of tubes.



To install the Sipper Module:

1. Draw the drain tube through the exit channel of the sipper from the inside outwards. The push-on connectors must be inside the sipper.
2. Draw the inlet tube through the entry channel of the sipper from the outside inwards. The push-on connectors must be inside the sipper.

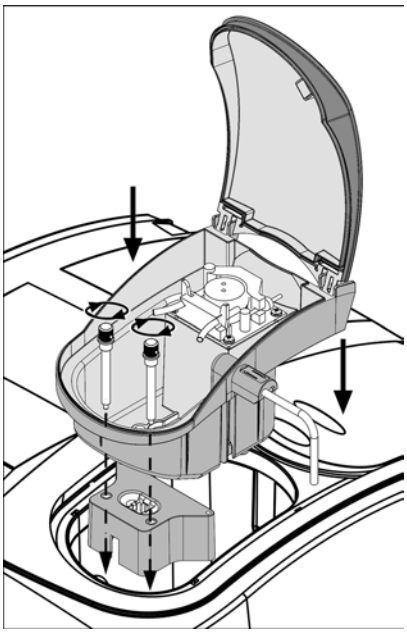
NOTE Take care to avoid causing any kinks in the tubes!

3. Turn the Sipper Module on its side.
4. Draw the inlet tube and the drain tube through, respectively, the entry channel and the exit channel of the sipper. The inlet tube must be drawn through the guide tube. The bottom end of the guide tube must click into the channel.

NOTE Take care to avoid causing any kinks in the tubes!

5. Hold the rubber fitting with the ridges over the grooves and push it firmly onto the guide tube and the waste tube.

6. Make sure the rubber fitting firmly encloses the guide tube and the drain tube.

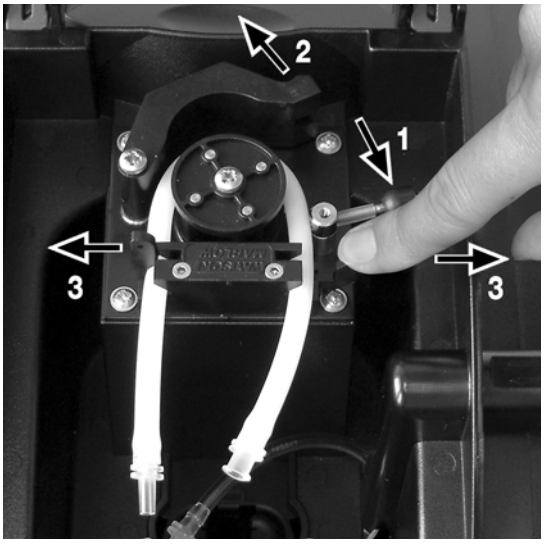


7. Open the cell compartment.

8. Without the locking screws, insert the Standard Cell Holder or Microcell Holder (see "Standard Cell Holder Installation" on page 14) in the cell compartment.

9. Place the Sipper Module on the Standard Cell Holder or Microcell Holder in such a way that the screw holes are positioned exactly one above the other. The lid of the Sipper Module can be opened towards the back of the instrument.

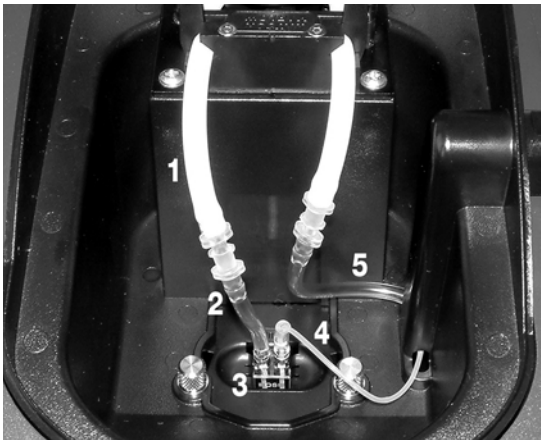
10. Secure the Sipper Module and the Standard Cell Holder or Microcell Holder with two locking screws (see Figure 3.13).



11. Pull the pump adjustment forward (1) and open the pump tubing clamp (2).

12. Wrap the white pump tubing around the pump and clamp the ends on the right and left in the front two retainers (3).

NOTE The push-on connectors of the pump tubing must be positioned as shown in the illustration.



13. Insert the flow cell (3).
14. Use the outlet connector (2) to join the white pump tubing (1) to the flow cell outlet (3).
15. Connect the drain tube (5) to the right end of the white pump tubing.
16. Connect the inlet tube to the inlet of the flow cell (4).

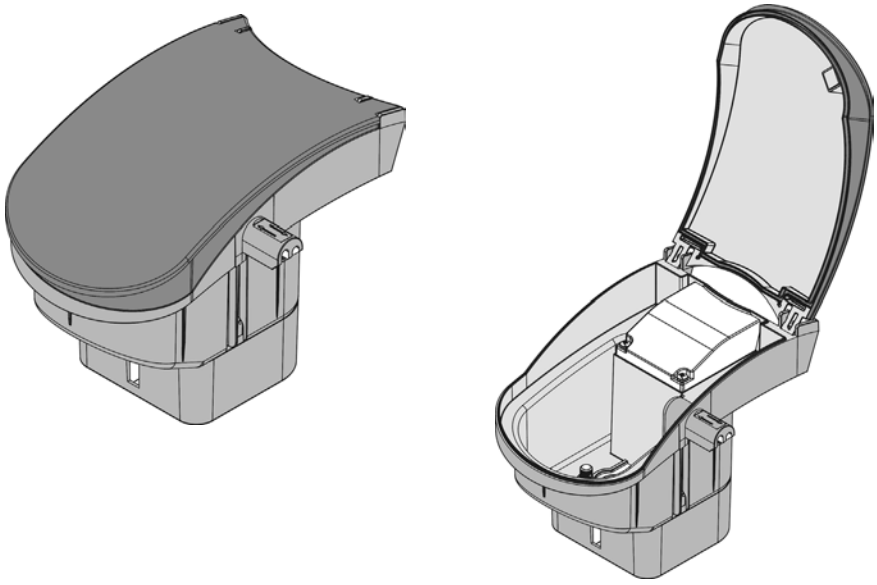
Peltier Temperature Control Module

The Peltier Temperature Control Module allows you to heat or cool the sample in the range of 15°C to 50°C (59°F - 122°F), set an alert for temperature deviation, select units of centigrade or Fahrenheit, and disable temperature control after a specified time.

You can set these parameters using Setup Mode.

Temperature control starts when you enter the temperature; it continues until either you or the idle parameter turns off the temperature.

Figure 3.15 Peltier Module



Peltier Temperature Control Module Installation

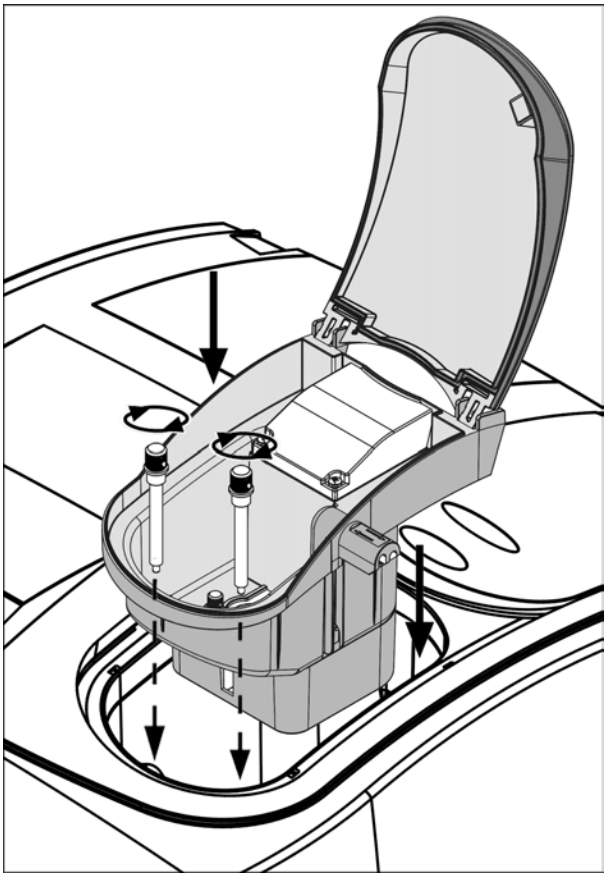
To assemble the Peltier Module:

1. Open the cell compartment.

2. Insert the Peltier Module in the cell compartment with the module lid open toward the back of the instrument (see Figure 3.16).

3. Secure the Peltier Module with two locking screws.

Figure 3.16 Installing the Peltier Temperature Control Module



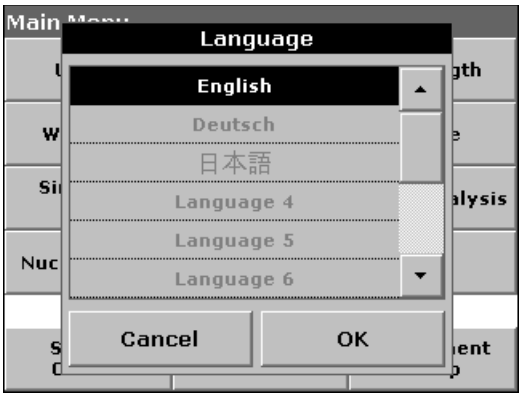
3.4 Switching the Power On

To switch on the instrument:

1. Close the cell compartment.
2. Press the power switch on the back.

WARNING Do not switch the instrument off and on in rapid succession. Always wait about **5 seconds** before switching the instrument on again; otherwise, you may damage the electronic and mechanical systems.

Language Selection

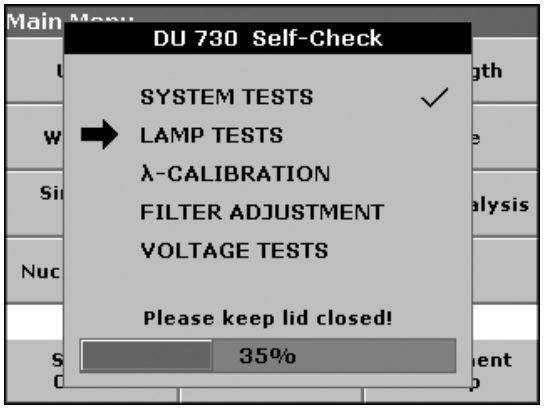


The software of the DU Series 700 Spectrophotometer includes several language options. To choose a language:

1. While turning on the instrument, touch the display until the screen displays the list of languages.
2. Select the desired language.
3. Touch **OK** to confirm the language selection. The self-check starts automatically.

NOTE The instrument continues to power up using the selected language, until you select a different language.

Self-Check



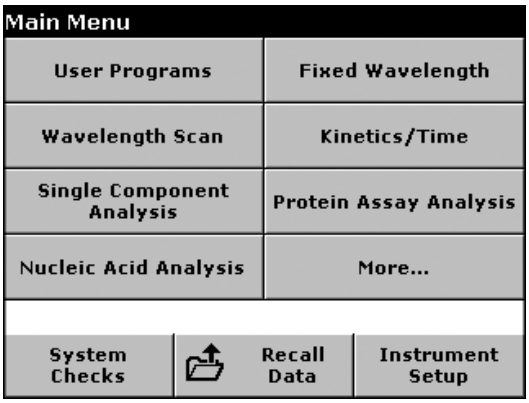
Each time you power up the instrument, it performs a series of diagnostic tests automatically to ensure operation of major system components.

This procedure, which takes approximately two minutes, checks the system, lamps, wavelength calibration, filter adjustment, and voltage. The display adds a check mark next to the test name to confirm its functionality.

When power-up diagnostics are complete, the Main Menu appears.

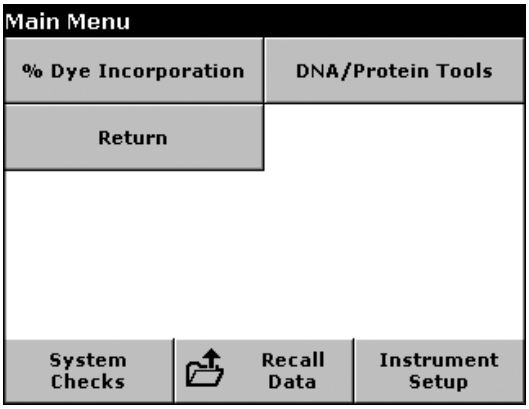
NOTE If any function does not pass or if any error messages appear during self check, see "Troubleshooting" on page 147.

3.5 Main Menu



The Main Menu consists of two pages on the DU 730. From there you can access common functions as well as all the available applications modes. Table 3.1 identifies each Main Menu soft key and briefly describes the corresponding operation mode.

- To display the second page of the Main Menu, touch **More...**.



- To return to the first page, touch **Return**.

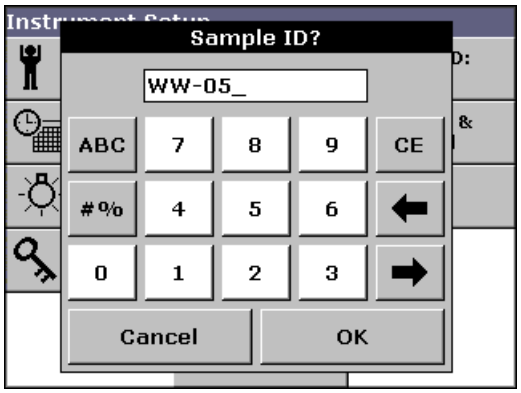
Table 3.1 Main Menu Soft Key Options

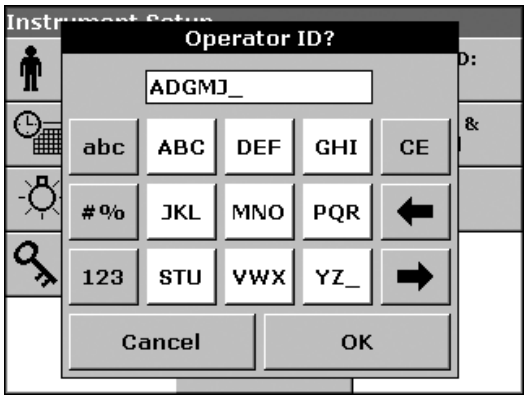
Soft Key	Operation Mode
User Programs	The User Program mode lets you recall saved programs.
Fixed Wavelength	The Fixed Wavelength mode collects data at up to 4 wavelengths. You can display the data in absorbance, transmittance, or concentration. The concentration is calculated by a user-defined factor or formula.
Wavelength Scan	The Scan Wavelength mode shows how a sample absorbs light over a range of wavelengths. You can use this feature to optimize instrument sensitivity with a given chemistry. The screen plots the scan.
Kinetics/Time	The Kinetics/Time Measurement mode records absorbance or % transmittance at a single wavelength over a specified time period.
Single Component Analysis (SCA)	The SCA mode lets you prepare a standard curve for analyzing samples that contain one component.
Protein Assay Analysis	The Protein Assay mode provides a simple procedure to calculate the amount of protein in a sample. It contains pre-selected parameters for seven common protein assay methods.

Table 3.1 Main Menu Soft Key Options

Nucleic Acid Analysis	The Nucleic Acid mode provides ten methods for DNA, RNA, and oligonuceotide analyses, which calculates nucleic acid absorbance ratios and concentrations. Other calculations include molecular weight, nucleotide length, extinction coefficient, concentration, and melting temperature.
% Dye Incorporation	The % Dye Incorporation mode works for single- and duo-color methods (spotted array). This works for any dye that gives similar fluorescence wavelength to Cy3 (550nm) or Cy5 (650 nm).
DNA/Protein Tools	The DNA/Protein Tools mode does not take measurements. The mode provides a number of calculations, conversations, and tables to aid you in common daily tasks.
System Checks	The System Checks mode lets you check the performance of the instrument (photometric accuracy, photometric noise, stray light, wavelength accuracy, lamp history, and printer check).
Recall Data	The Recall Data mode lets you store, recall, send, and erase data from the Datalog.
Instrument Setup	The Instrument Setup mode allows you to configure the instrument and the module, based on your specific requirements.

3.6 Alphanumeric Keypad





Each time an operation requires input, the alphanumeric keypad opens. Use this screen to enter letters, numbers, and symbols as needed when programming the instrument. Unavailable options are grayed out, indicating that those functions do not apply to your operation mode.

The icons to the left of the screen allow you to choose an entry mode:

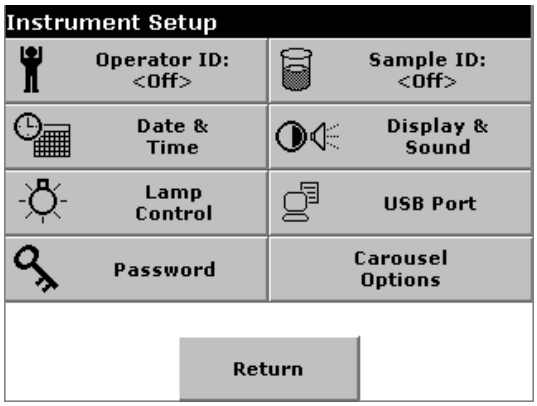
- **ABC (alphabetic):** When entering alphabetic characters (such as user-entered units), this key allows you to toggle between upper and lower case letters. It is inactive on some screens.
- **#% (symbols):** Lets you enter punctuation, symbols, and numerical subscripts and superscripts after touching this key.
- **123 (numeric):** Use this key to switch to a numeric keypad when you need to enter regular numbers.

The central part of the keypad changes to reflect the chosen entry mode. To enter a character, you must repeatedly touch a key until the desired character appears on the screen. To enter a space, use the underscore on the **YZ_** key.

The keys to the right side of the screen are:

- **CE** (clear entry): Clears all text displayed in the entry field.
- Left Arrow (backspace): Moves the cursor back one position, deleting the previous character.
- Right Arrow (advance): Advances the character to display the next one when the key shows more than one character.

3.7 Instrument Setup Mode

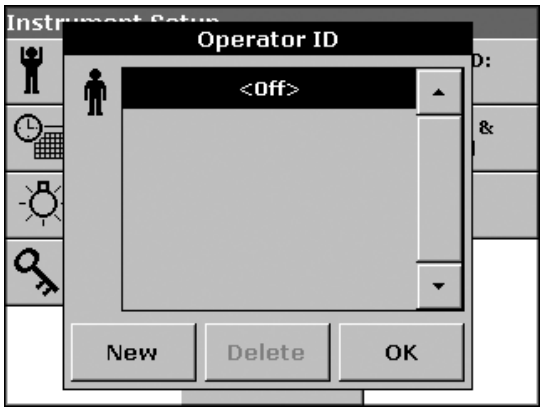


To change to Instrument Setup Mode, select **Instrument Setup** in the Main Menu.

The Instrument Setup menu displays a list of options you can use to set instrument functions.

If the instrument recognizes a Sipper module or a Peltier module, the **Carousel Options** key is replaced by **Sipper Options** or **Peltier Options**.

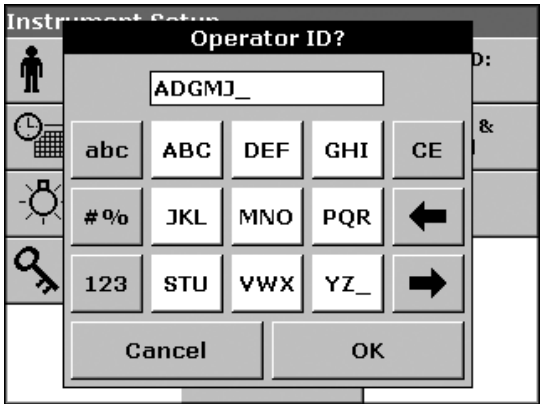
Operator ID



Use the Operator ID screen to enter up to 30 sets of operator initials (up to five characters each) into the instrument. You can assign the Operator ID to a measurement from the measurement screen or the **Operator ID** menu. This feature helps record which operator measured each sample.

To create an Operator ID:

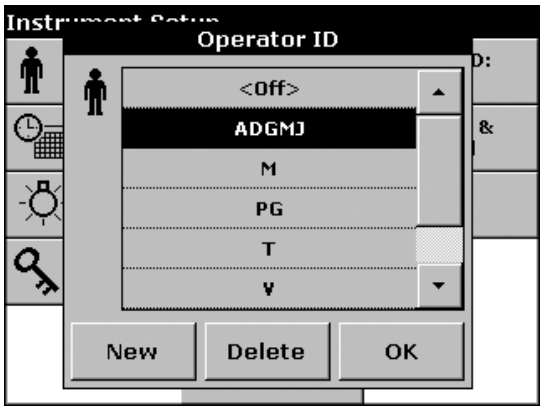
1. Touch **Operator ID** in Instrument Setup mode.
2. Touch **New** to enter a new **Operator ID**.



3. Use the alphanumeric keypad to enter a new Operator ID (see "Alphanumeric Keypad" on page 30).

NOTE Use the alphanumeric keypad to enter a new Operator ID. You cannot use spaces in this function. Use underscore symbols instead (**YZ_** key).

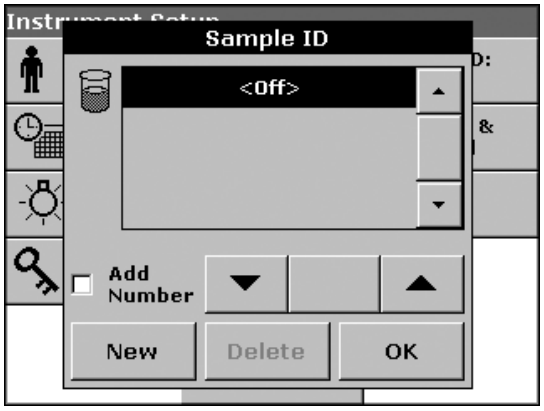
NOTE Touch **Delete** to remove an Operator ID from the list.



4. Touch **OK** to save your changes. The screen returns to the Instrument Setup menu and shows the selected operator identifier.

NOTE If an Operator ID is active, you can also touch the Operator ID icon directly on the Measurement screen. The Operator ID screen opens, in which you can change the ID.

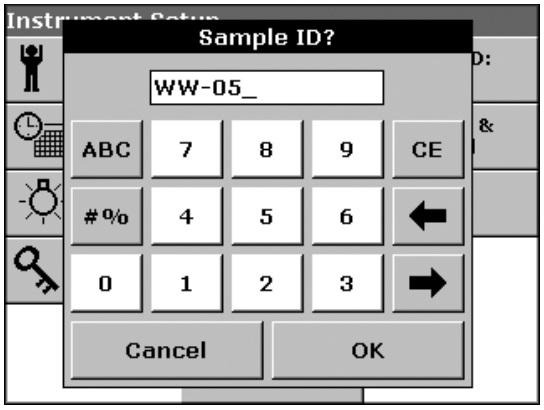
Sample ID



Use the Sample ID screen to enter up to 30 sample identification tags (up to 13 characters each) into the instrument. You may want to use this to identify the samples more effectively.

To access this function:

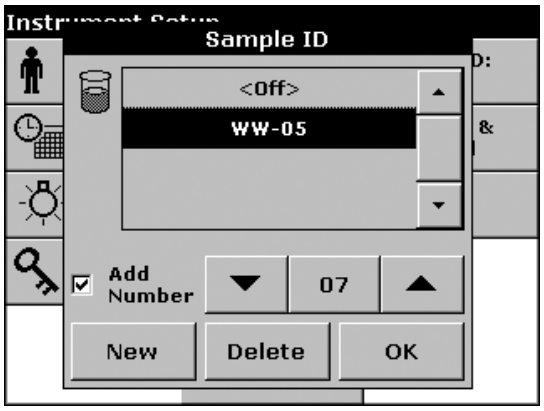
1. Touch **Sample ID** in the Instrument Setup menu.
2. Touch **New** and enter a new **Sample ID**.
   - If you want to subdivide the Sample ID with numbers, touch **Add Number**.
   - Use the arrow keys to specify how many numbers can be added for the sample identification.
   - Touch the key between the arrow keys to add directly the number of choice.
   - Use the alphanumeric keypad to enter a new Sample ID (see "Alphanumeric Keypad" on page 30).



NOTE You cannot use spaces in this function. Use underscore symbols instead (**YZ_** key).

3. Touch **OK** to confirm. The display shows the chosen Sample ID, as shown in the following illustration.

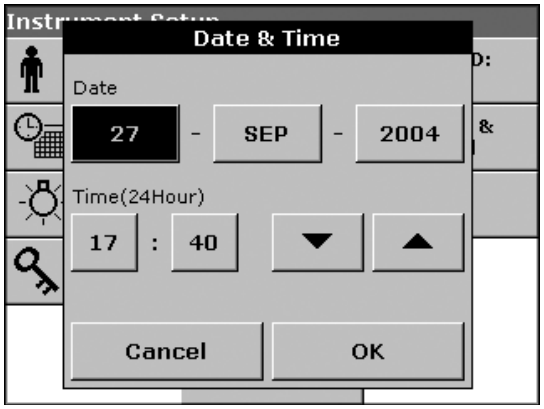
NOTE Touch **Delete** to remove a Sample ID from the list.



4. Touch **OK** to save your changes. The screen returns to the Instrument Setup menu and shows the selected sample identifier.

NOTE If a Sample ID is active, you can also touch the Sample ID icon directly on the Measurement screen to change it. The Sample ID screen opens, in which you to change the Sample ID.

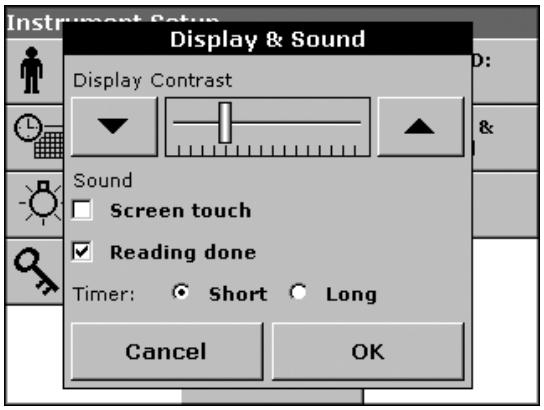
Date and Time



Use the Data & Time screen to set the instrument's data and time.

1. Touch **Date & Time** in the Instrument Setup menu. The screen provides the fields necessary to set the date and time.
2. Touch the appropriate field and use the arrow keys to change the value.
3. Touch **OK** to confirm. The screen returns to the Instrument Setup menu.

Display and Sound Preferences



Use the Display & Sound screen to set the audiovisual preferences.

1. Touch **Display & Sound** in the Instrument Setup menu. The screen displays four options:
   - **Display/Contrast:** lets you adjust the display contrast to suit lighting conditions and viewing angle.
   - **Screen touch:** The instrument default is off. To make a short beep every time by touching the screen activate the **Screen touch** option.
   - **Reading done:** The instrument default is set to make a short beep every time a reading is complete. To turn off the Reading done sound, deactivate the **Reading done** option.
   - **Timer:** lets you change the length of the timer sound touch **Short** or **Long**. Long beeps are better for noisy environments.
2. Touch **OK** to confirm. The screen returns to the Instrument Setup menu.

Lamp Control

The halogen gas-filled tungsten lamp produces light in the visible spectrum 320 to 1100 nm.

The deuterium lamp (UV-lamp) produces light in the ultraviolet spectrum 190 to 360 nm.

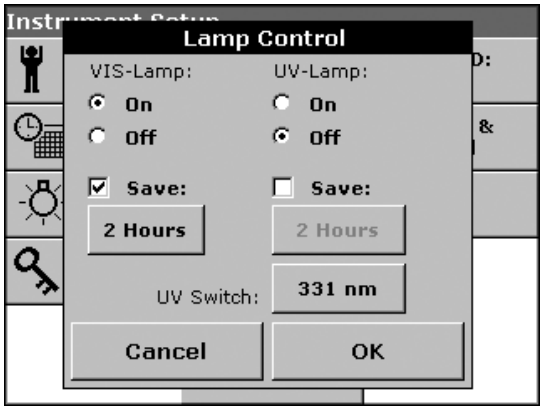
In the overlap zone from 320 to 360 nm, you can use either the deuterium lamp (UV-lamp) or the halogen lamp for measurements.

The lifetime of the lamps is influenced by on-off operation and the length of time in use.

Typically, you might turn the instrument on for the entire 8-10 hour shift, and then off until the next day.

NOTE In general, avoid on/off cycles of the lamp; this shortens the lamp's life span. For maximum life, turn the lamp off only if it will remain off for at least 4-5 hours.

If the instrument needs a lamp for the selected program, or the instrument is operating inside the lamp's spectrum, the lamp switches on automatically.



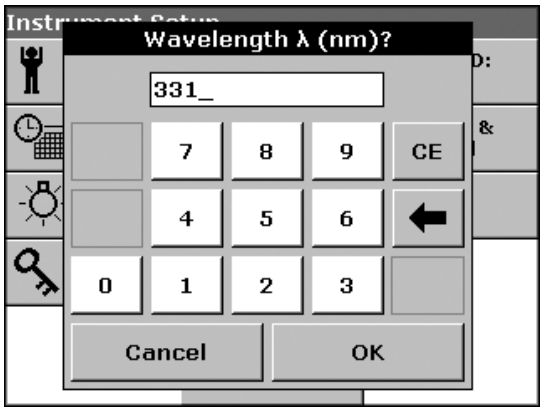
Use the Lamp Control screen to set the lamp timer switches.

1. Touch **Lamp Control** in the Instrument Setup menu.
2. Select **On** to switch on the VIS-Lamp.
3. To select the time for automatic lamp shutoff:
   a. Select the **Save** check box, located below the VIS-Lamp or UV-Lamp control option. Depending on your selection, the screen displays either the Visible Lamp Save screen or UV Lamp Save screen.

   b. Select the length of time you want the lamp switched on.
   c. Touch **OK**.

NOTE After this period of time the lamp automatically turns off under one of these conditions:

- when no activity has taken place or
- when the measurements have been outside from the lamp's wavelength range.

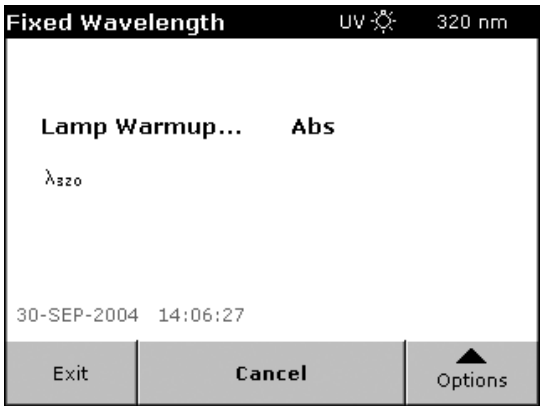


4. If necessary, touch the UV Switch button to select a wavelength between 320 and 360 nm, at which the instrument changes from the visible to the UV source. Use the alphanumeric keypad to enter the switching wavelength.

NOTE Usually, there is no need to change the default value of 331 nm for the switching wavelength.

5. Touch **OK** to confirm. The screen returns to the Instrument Setup menu.

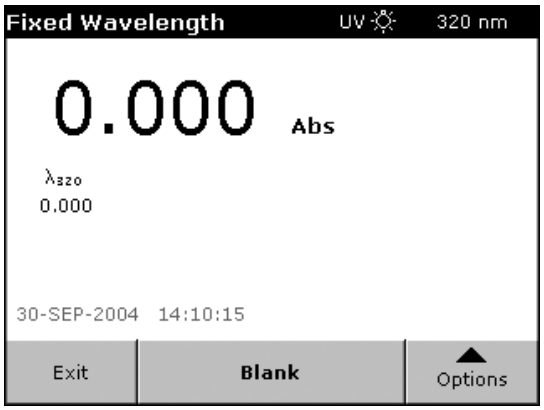
NOTE A program that requires a lamp has the highest priority. If, for example, the lamp in the Lamp Control screen was turned off but the current program requires the lamp, the lamp turns on automatically.



Measuring Mode Effects:

1. UV-Lamp switches on manually. The UV Lamp icon appears flashing in the display.
2. The instrument displays “Lamp Warmup...”

NOTE For the most accurate and stable measurements in the UV range, let the lamp warm up for five minutes before taking the first reading.



3. When the lamp is warmed up and ready, the UV icon stops flashing.

NOTE If both lamps are **On**, the symbol UV-VIS appears in the selected Measurement Mode.

NOTE As an alternative, you can touch the UV-Lamp icon on the measurement screen to change the actual setting.

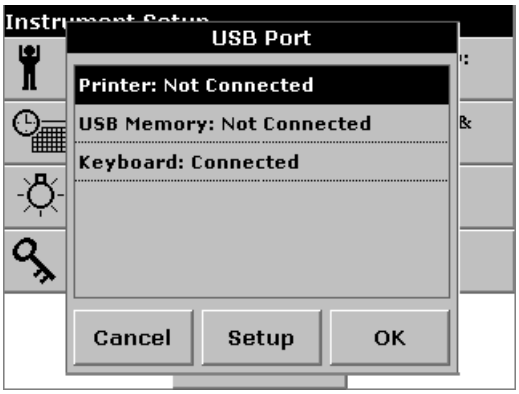
USB Port

As a standard feature, the instrument has three ports, located on the back wall of the instrument (Figure 2.3). These ports enable you to output data and graphics to a printer. They also let you connect a USB keyboard or a memory device.

Table 3.2 Connector Ports

Connector	Description
USB 2 and 3 ("A" Connectors)	These two USB ports let you connect a USB printer, a keyboard, a memory device, or a device for memory cards. These devices are controlled by the spectrophotometer.

NOTE The printer must have a USB interface and must feature the Printer Command Language (PCL). Printers that use the Lightweight Imaging Device Interface Language (LIDIL) or are based on the Printing Performance Architecture (PPA) require Microsoft Windows and cannot be used with a DU Series 700 instrument.



To view a list of devices connected to the ports:

1. Touch **USB Port** in the Instrument Setup menu. The USB Port screen displays a list with information about the following connections:

   • Printer

   • USB Memory

   • Keyboard

NOTE This screen provides information only. The instrument automatically detects connected devices.

2. Touch **OK** to return to the Instrument Setup menu.

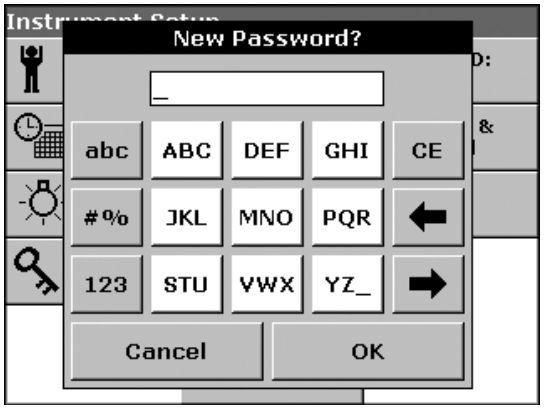
Password

The Password screen enables you to set a password and a variety of security settings. The security list enables you to control the accessibility of various functions. For example, you can prevent changing or deleting different stored programs without authorization.

To access the security settings:

1. Touch **Password** in the Instrument Setup menu.
2. Touch **Set Password.**

NOTE This enables you to assign a password required for the specified function. You must assign a password before the screen activates the **Security List** option.

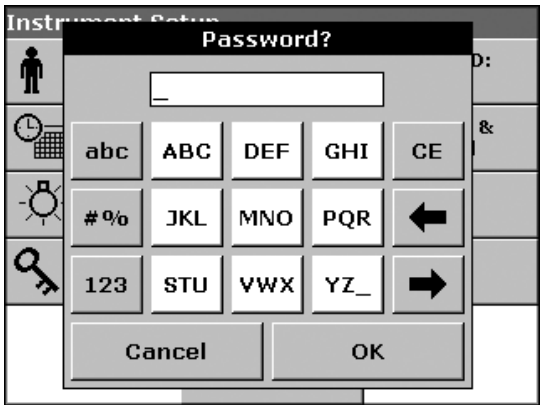


3. Use the alphanumeric keypad to enter a new password.
4. Touch **OK** to confirm the entry. This activates the Security List.

5. Touch **Security List** to lock various functions from unauthorized users.

6. Check the functions you want to control by touching each respective option.
7. Touch **OK** to confirm the security options selected from the Security List.
8. Enter the new password again to confirm.
9. Touch **OK** to return to the Instrument Setup menu.

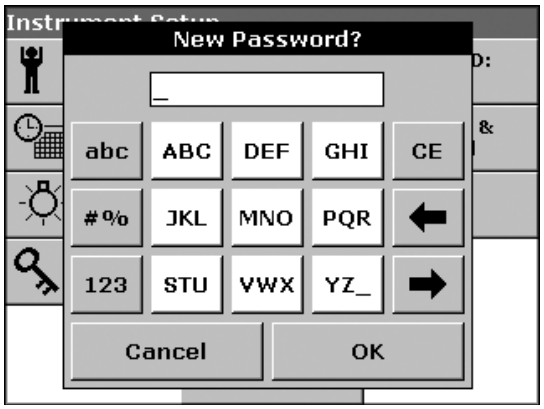
NOTE Each time a user attempts to perform one of the locked functions, an alphanumeric keypad opens, in which the user must enter the password before using those locked functions.



To deactivate a password:

1. Touch **Password** in the Instrument Setup menu.
2. Use the alphanumeric keypad to enter the former password and confirm with **OK**.

3. Touch **Set Password**. This opens the alphanumeric keypad.

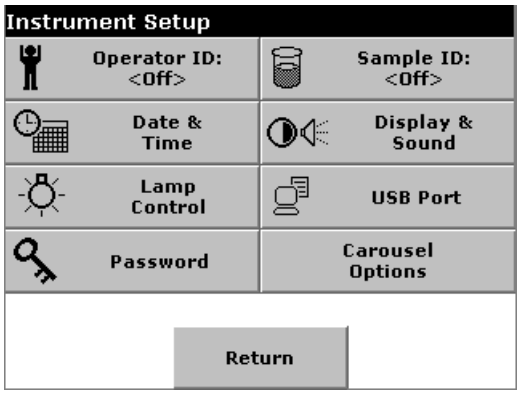


4. On the alphanumeric keypad, leave the New Password field blank and touch **OK**. This deactivates the former password and returns to the Password screen.

NOTE Use this function to delete the former password or to enter a new one.

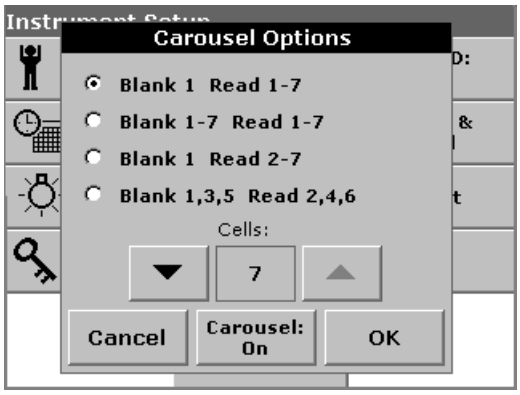
Carousel and Module Options

Carousel



If your instrument includes a Carousel Holder, the Instrument Setup menu includes **Carousel Options**. You can activate the Carousel Holder and modify the carousel parameters using in this menu.

1. Activate the Carousel Holder:
   a. Touch **Carousel Options** in the Instrument Setup menu.
   b. Touch **Carousel: Off** to change its display to **Carousel: On**.
   c. Touch **OK** to confirm your selection.

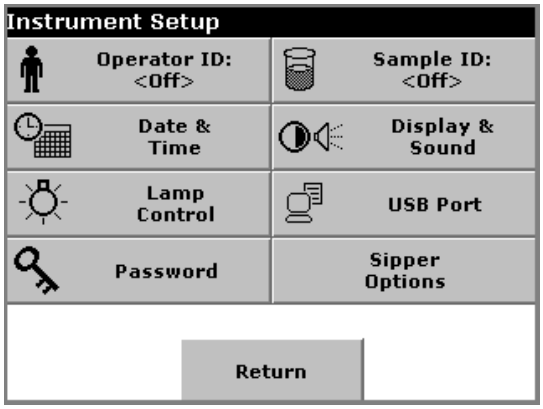


2. Set the carousel options:
   a. Touch **Carousel Options** and touch
   b. Select the carousel option of your choice.

      For example, the first option is **Blank 1 Read 1-7**, which means that the blank cuvette will be read in position 1 while samples must be placed in the cell positions 1-7. With this option, you must remove the blank cuvette after blanking.
   c. Use the arrow keys to specify the number of cell positions you want to use.
3. Touch **OK** to confirm and return to the Instrument Setup menu.

NOTE As an alternative, you can access the Carousel Options screen directly from the measurement mode by touching the **Carousel** Icon on the bottom-right of the screen. The Carousel Options screen appears and you can quickly make the necessary changes.

Sipper Module



If your instrument includes a Sipper Module, the Instrument Setup menu provides **Sipper Options**. You can modify the sipper parameters using this menu.

1. Touch **Sipper Options** in the Instrument Setup menu to display the Sipper Options screen.

2. Modify the desired parameters for the following options:

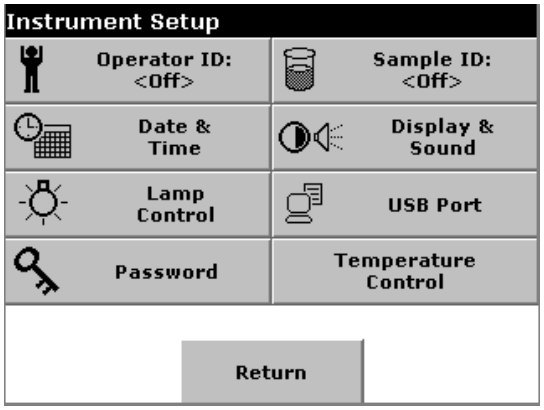
   - **Sip Time:** The sip time, in conjunction with the pump adjustment, determines the amount of sample delivered to the sample cell. Enter the amount of time (1-99 seconds) for the pump to run to aspirate the sample into the flow cell.

   - **Settle Time:** The settling time defines how long the instrument waits between turning off the pump and taking a sample reading. Use this interval to allow bubbles and sample turbulence to settle out of the light path. Enter the amount of time (1-99 seconds) the instrument waits after the sip time before taking a sample reading. During this time, the sample stops flowing and stabilizes.

   - **Purge Time:** The purge time, in conjunction with the pump adjustment, determines the amount of air or rinsing solution pulled through the sample cell after a reading. This cycle is optional; you can program the Sipper to recover the sample or to send the sample to a waste vessel. Enter the amount of time (1-99 seconds) for the pump to run to empty the sample from the flow cell after the sample reading.

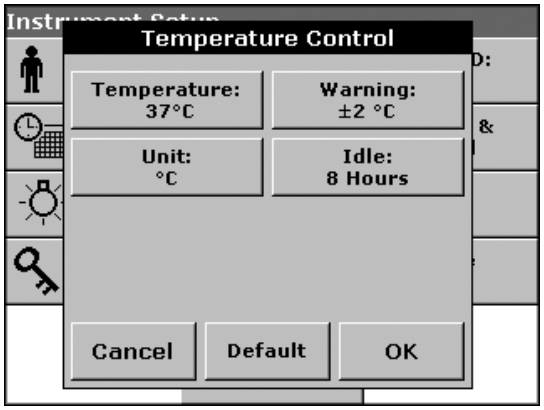
   - **Purge Start: Auto/Manual**: If **Purge** is enabled, use this option to toggle between **Manual** and **Auto**. In **Manual** mode, you must initiate the sample purge. In **Auto** mode, the module automatically purges the sample as soon as a reading is taken.

Peltier Module (Temperature Control)



If your instrument includes a Peltier Temperature Control Module, the Instrument Setup menu provides a **Temperature Control** option. You can modify Temperature Modes using this menu.

1. Touch **Temperature Control** in the Instrument Setup menu to display the Temperature Control screen.



2. Modify the desired parameters for the following options:

   - **Temperature:** Turns temperature control off or on and allows you to set the desired temperature.
   - **Warning:** The warning alerts you when the actual temperature deviates from the set temperature. You can turn the warning off or to on by entering the deviation from set temperature that triggers the warning.
   - **Unit:** Select the temperature unit as Centigrade or Fahrenheit.
   - **Idle:** The idle function causes the instrument to automatically turn temperature control off after the specified time. Use this button to turn the idle function off or on by entering the amount of time in hours before the temperature control is automatically turned off.

NOTE As an alternative, you can access the Temperature Control screen directly from the measurement mode by touching the Temperature Control icon at the bottom-right of the screen. The Temperature Control screen appears and you can quickly make the necessary changes.

User Programs

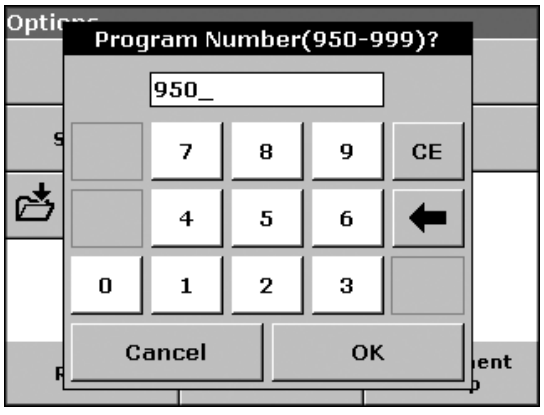
The User Program mode provides access to programs or modes that have been defined by the user(s). A user program is one of the application modes with customized parameters. The instrument can store up to 50 user programs.

4.1 Storing a User Program

After selecting the analysis parameters in many off the modes, you can store the parameters for later recall.

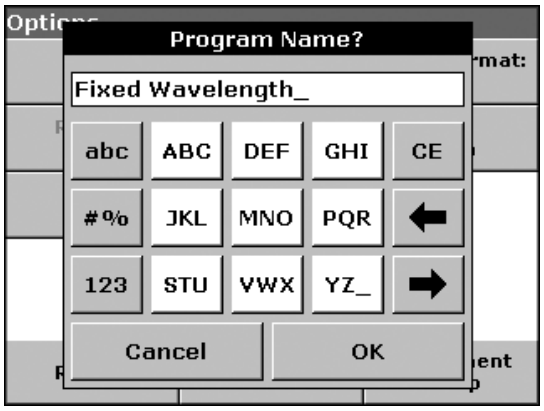
NOTE Wavelength Scan and Kinetics/Time parameters cannot be saved as a User Program.

1. Set the desired analysis parameters in the mode of your choice.
   - **Fixed Wavelength**: Touch **Options** then **More...** (see "Parameter Setup" on page 45).
   - **Single Component Analysis**: Touch either **Standard Curve** or **Coefficients** (see "Parameter Setup" on page 61).
   - **Protein Assay Analysis**: Touch **Start**, then either **Standard Curve** or **Coefficients** (see"Parameter Setup" on page 71).
   - **Nucleic Acid Analysis**: Touch **Start**, **Options**, then **More...** (see "Parameter Setup" on page 81).
   - **% Dye Incorporation**: Touch **Options** then **More...** (see "Parameter Setup" on page 100).



2. Touch **Save as User Program**.
3. Enter a user Program Number between 950 and 999 and touch **OK**.

NOTE If you enter a program outside this range, the instrument beeps and displays the next available number. If you enter a Program Number that is currently in use, confirm whether you want to replace it.

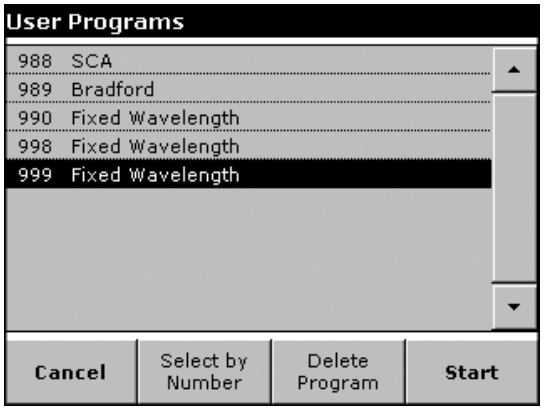


4. In the Program Name screen, enter a Program Name with up to 28 characters.

NOTE The alphanumeric keypad appears with a default name (name of the mode) for the user program. You can use the keypad to change the name or enter a different name.

5. Touch **OK** to save the program parameters under the chosen name and return to the operating mode.

4.2 Recalling a User Program



To recall a previously stored user program:

1. Touch **User Programs** in the Main Menu to display all available user programs.
2. Select the user program: touch the respective line and highlight the program or touch **Select by Number** and enter the User Program Number, then touch **OK**.
3. Touch **Start** to run the program.

4.3 Deleting a Stored User Program

To delete a previously stored user program:

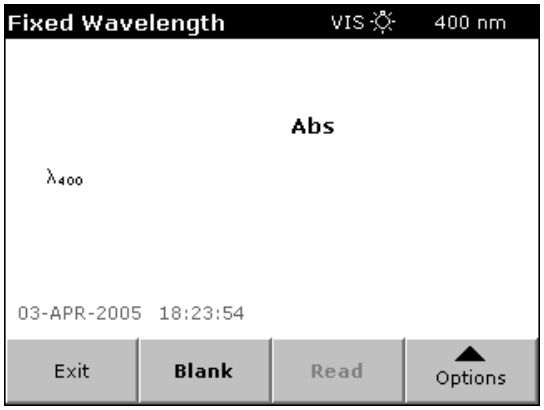
1. Touch **User Programs** in the Main Menu to display a list of all user programs.
2. Select the user program: touch the respective line and highlight the program or touch **Select by Number** and enter the User Program Number, then touch **OK**.
3. Touch **Delete Program.**
4. Touch **OK** to confirm deletion.

Fixed Wavelength Mode

The Fixed Wavelength mode lets you collect data at up to 4 wavelengths. You can take and display the data in absorbance or transmittance and calculate the concentration of your samples using a factor or a formula.

- **Absorbance** measures the amount of light absorbed by the sample, in units of Absorbance.
- **% Transmittance** measures the percent of the original light that passes through the sample and reaches the detector.

5.1 Parameter Setup



To enter the Fixed Wavelength mode, touch **Fixed Wavelength** in the Main Menu.

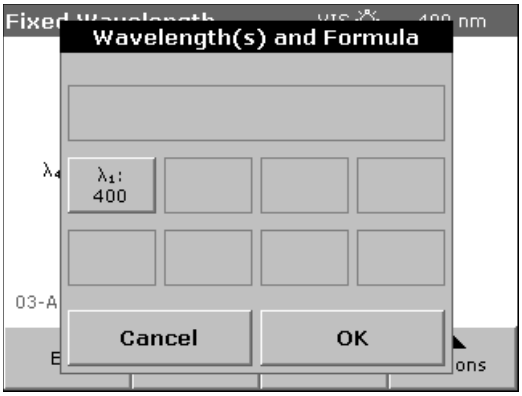
Touch **Options** for Parameter Setup. This screen provides the following options:

- **More...:** displays more options in the Options screen.
- **Store Off/On:** lets you toggle the storage setting from **Off** to **On** and manually store data.
- **% Trans/Abs:** toggles between %Trans (Transmittance) and Abs (Absorbance) display/readings.

NOTE **Abs** is in place of **%Trans** after you have switched to % Transmittance.

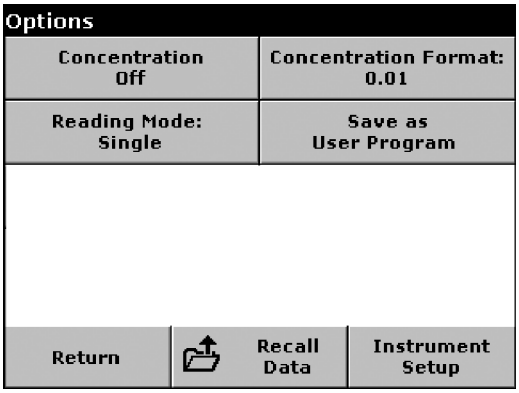
- **Timer icon:** sets the timer. Using the timer does not affect the readings that the instrument is taking. The times are not stored with sample readings.
- **λ:** lets you enter the wavelength settings.

NOTE The following sections describe how to set the wavelength(s), the formula, and the coefficients or factors.



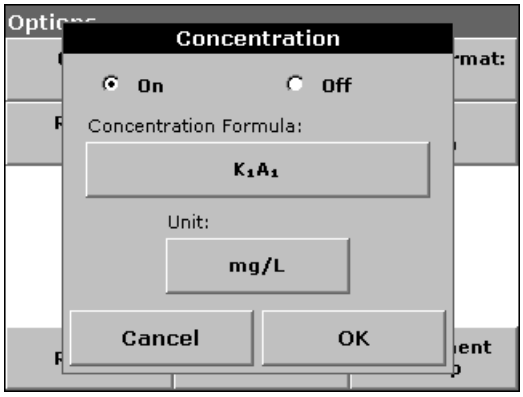
Touch λ to enter the desired wavelength.

NOTE For more information on how to set the wavelength(s), the formula, and the coefficients, or factor, see "" on page 47.



Touch **More…** to go to the Options screen. This screen provides the following options:

- **Concentration (Off/On):** allows you to switch the concentration calculation on or off. Also, you can choose the a formula and determine factor(s) for up to 4 wavelengths to calculate the concentrations.
- **Concentration Format:** sets the number of significant digits for the calculated concentration.
- **Reading Mode:** toggles between **Single** and **Continuous** readings.
- **Save as User Program:** stores the selected parameters as a user program (see "User Programs" on page 43).
- **Recall Data:** see "Recalling Data" on page 127.
- **Instrument Setup:** see "Instrument Setup Mode" on page 31.



The Concentration screen lets you switch the concentration calculation on or off.

When turned **On**, the Concentration Formula button becomes enabled and you can touch it to access the Concentration Formula screen, which allows you to select a formula. The factor(s) for the concentration calculation are also determined in this screen, together with the λ selection.

Touch the **Unit** button to select a concentration unit or add a new unit to the pre-defined units. You may delete only user-defined units.

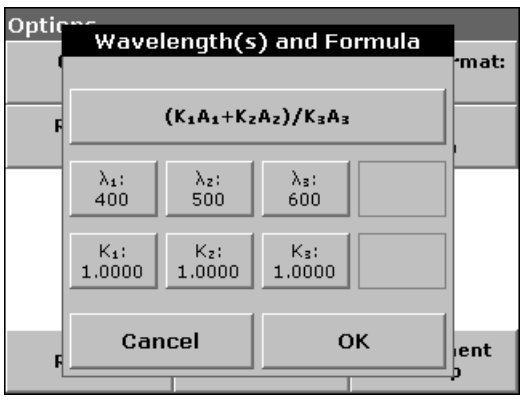
NOTE The selected unit is not shown unless a concentration is calculated (in this case, the factor must be other than 1.0000 and/or a formula other than $_{K1\ A1}$ has been selected).

Touch **OK** to save the parameter setup.

The following formulas are available:

$K_1 A_1$
$K_1 A_1 + K_2 A_2$
$K_1 A_1 + K_2 A_2 + K_3 A_3$
$K_1 A_1 + K_2 A_2 + K_3 A_3 + K_4 A_4$
$K_1 A_1 / K_2 A_2$
$(K_1 A_1 + K_2 A_2) / K_3 A_3$
$(K_1 A_1 + K_2 A_2) / (K_3 A_3 + K_4 A_4)$

A_1 refers to the absorbance at wavelength 1,
A_2 refers to the absorbance at wavelength 2, etc.
K_1 refers to the coefficient at wavelength 1,
K_2 refers to the coefficient at wavelength 2, etc.
Coefficients can be set negative where subtraction is required.

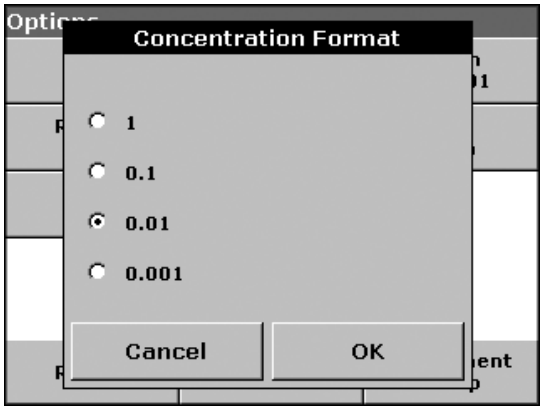


- To change the formula, touch the button that displays the current formula.
- To change a wavelength, touch one of the **"λx:"** keys.
- To change a coefficient, touch one of the **"K_X:"** keys.

Enter the desired wavelength(s) and/or coefficient(s) with the numeric keypad and touch **OK**.

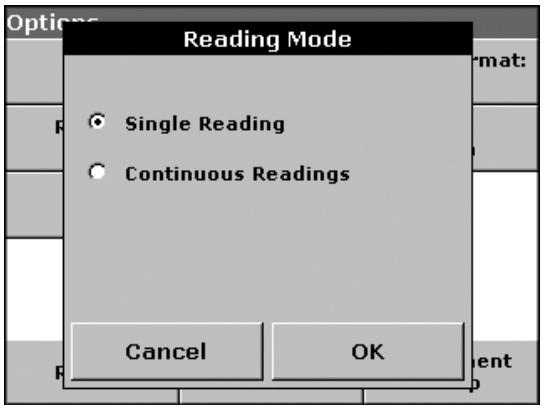
NOTE The instrument allows you to enter up to 5 significant digits, with a maximum of 4 significant digits after the decimal point.

Touch **OK** to save your selections.



To set the Concentration Format:

1. Touch **Concentration Format** in the Options menu.
2. Select the number of significant digits for the concentration results by touching the respective radio button.
3. Touch **OK** to confirm.



To select the reading mode:

1. Touch **Reading Mode** in the Options screen and choose one of these options:

   - **Reading Mode: Single**
     This feature is available for procedure readings to stop measurement reading fluctuations on the display once it reaches a stable reading. This is the default setting.
   - **Reading Mode: Continuous**
     This feature is only available by choosing **one** measuring wavelength.

NOTE With the continuous reading mode, only the **Blank** button is displayed. The **Read** button is not available since the readings are continuous and start automatically.

2. Touch **OK** to confirm your selection.

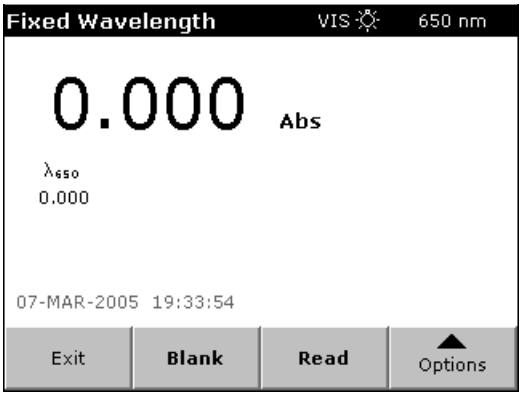
Analysis of Samples

After entering the analysis parameters, you are ready to read the samples.

This description assumes that you have selected **Single Reading** (default) as the reading mode and that you are using a cell holder that holds one cell (the Standard Cell Holder that comes with the instrument). For instructions on using this function with a Carousel Cell Holder, the Sipper Module, or the Peltier Temperature Control Module, see "Carousel, Sipper, and Peltier Operation" on page 117.

Before taking measurements, you need to blank the instrument on air (no cuvette in the light path) or by using a cuvette with a "blank" solution, which is recommended for best results.

NOTE **Read** is disabled until the blank reading is taken.



1. Insert the blank cuvette into the cell holder and close the cell compartment.
2. Touch **Blank** to take the blank reading. When done, the instrument enables the **Read** button.
3. Insert a sample cuvette into the cell holder and close the cell compartment.

NOTE If the lid of the cell compartment is open, the instrument disables the **Blank** and **Read** buttons.

4. Touch **Read** to take the sample reading.

When trying to read a Blank or a Sample, the appropriate source must be turned on. The system assures that this is the case. You may observe the following:

- The UV lamp automatically turns on after you have entered a wavelength in the UV spectrum. In this case, you have to wait for the UV lamp to warm up before you can take readings.
- During the warm-up phase of the UV lamp, the screen displays a "Warming up..." message and flashes the UV lamp symbol. When the UV lamp is turned on automatically, the instrument performs the reading as soon as the UV lamp is ready.

When finished reading the sample, the instrument displays the result (Absorbance Reading or Calculated Concentration).

NOTE For instructions on using a Carousel Cell Holder, a Sipper, or a Peltier Temperature Controller, see "Carousel, Sipper, and Peltier Operation" on page 117.

NOTE For instructions on printing and storing data, see "Store, Recall, Send, and Delete Data" on page 125.

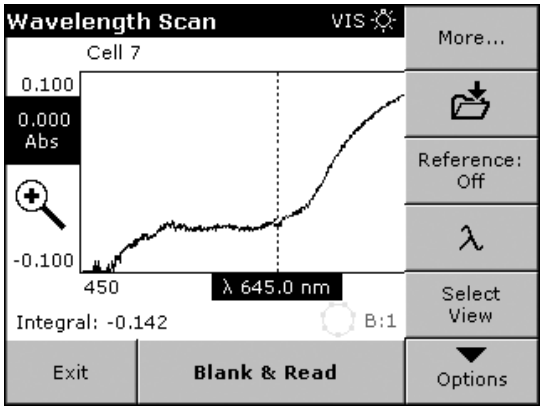
Wavelength Scan Mode

The Wavelength Scan mode shows how light is absorbed by a solution over a given wavelength range. You can display wavelength scans in either Absorbance (Abs) or Transmittance (%T). The scans can be scaled manually and automatically. This mode lets you determine peaks and valleys and zoom into areas to see more details.

You can obtain the absorbance or transmittance values at a given wavelength by moving the cursor to any point on the x-axis of the graph (Track function). You can also tabulate the scan data.

When recalling a stored scan, you can overlay this reference scan with the current scans.

6.1 Parameter Setup



To enter the Wavelength Scan mode:

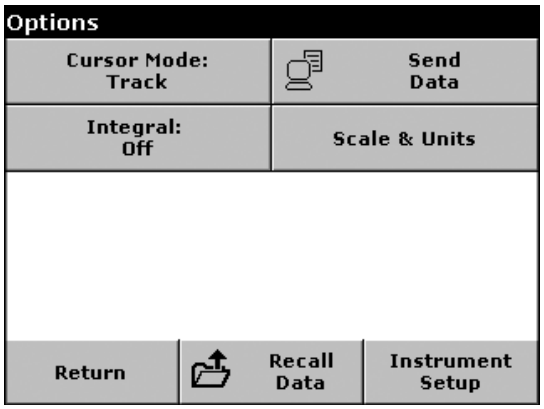
1. Touch **Wavelength Scan** in the Main Menu.
2. Touch **Options** for Parameter Setup.

   - **More...:** displays further options in the Options screen.
   - **Store Icon (Folder with Arrow):** stores the scan data.
   - **Reference (Off/On):** allows you to select a reference scan from the stored scans and overlay the current scans with the reference.

NOTE This option is available only when there are stored scans and the selected reference has the same wavelength range and steps.

   - **λ:** lets you enter the wavelength range (lower and upper limit as well as the scan resolution or step).
   - **Select View:** toggles between Graph (scan) and Table (tabular data).

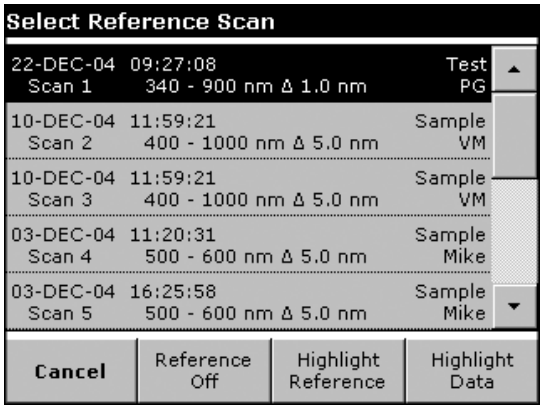
NOTE The screen displays **Select View** after the first scan.



3. Touch **More...:** to go to the previous page of the Options screen.

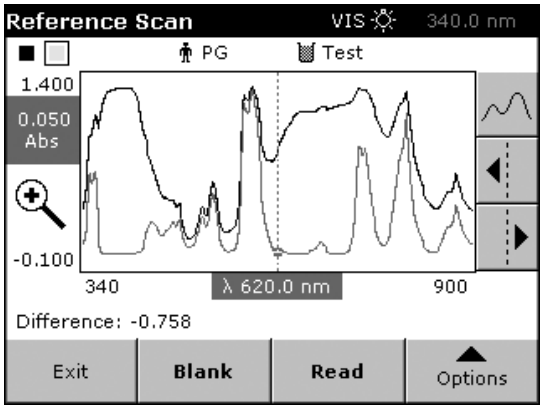
   - **Cursor Mode:** lets you select **Track** or **Peak/Valley**. With **Track**, the cursor moves to the next or previous step. With **Peak/Valley**, the cursor moves to the next or previous peak or valley.
   - **Send Data:** sends data to a printer, computer, and storage device.
   - **Integral (On/Off):** when toggled to **On**, the scan integral is displayed in the bottom-left corner of the screen, replacing the displayed date.
   - **Scale & Units:** lets you select **Absorbance** or **%Transmittance** and determine the y-axis scaling (manual or automatic).

NOTE For instructions on storing and printing scan data, see "Store, Recall, Send, and Delete Data" on page 125.


Select Reference Scan
22-DEC-04 09:27:08 Test
Scan 1 340 - 900 nm Δ 1.0 nm PG
10-DEC-04 11:59:21 Sample
Scan 2 400 - 1000 nm Δ 5.0 nm VM
10-DEC-04 11:59:21 Sample
Scan 3 400 - 1000 nm Δ 5.0 nm VM
03-DEC-04 11:20:31 Sample
Scan 4 500 - 600 nm Δ 5.0 nm Mike
03-DEC-04 16:25:58 Sample
Scan 5 500 - 600 nm Δ 5.0 nm Mike
Cancel
Reference Off
Highlight Reference
Highlight Data

To recall stored scan data to be used as a Reference Scan for overlays:

1. From the Wavelength Scan mode, touch **Options**.
2. Touch **Reference: Off**.
3. In the Select Reference Scan screen, touch the line with the desired Scan # to highlight it.
4. Touch **Highlight Reference** or **Highlight Data**.



The system returns to the scan mode, showing the Reference Scan as an overlay, if a scan has been taken already. The button in the Options menu now indicates **Reference: On**.

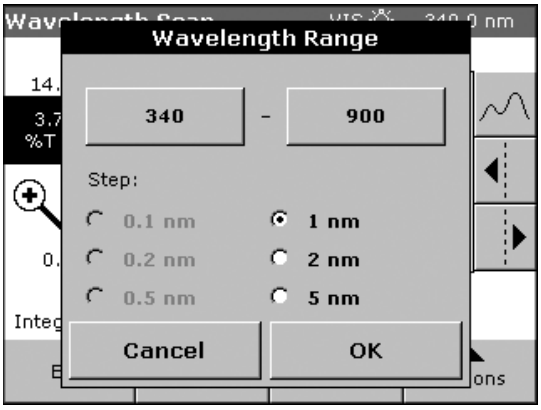
The Reference Scan is shown in gray and the Measured Scan in black.

NOTE This option can display only scans that have the same wavelength range and resolution.

A black and a gray box in the top-left corner of the screen identifies the scan for which the cursor is active.

Touch the black or gray box in the top-left corner to switch the cursor from the measured scan to the reference scan and vice versa.

The bottom-left corner of the display shows the Difference between the two scans in absorbance or transmittance.



To set the scan or wavelength:

1. Touch the λ key in the Options menu.
2. Touch the top-left button to change the lower limit and enter the desired value using the numeric keypad.
3. To set the upper limit, touch the top-right button and enter the desired value using the numeric keypad.

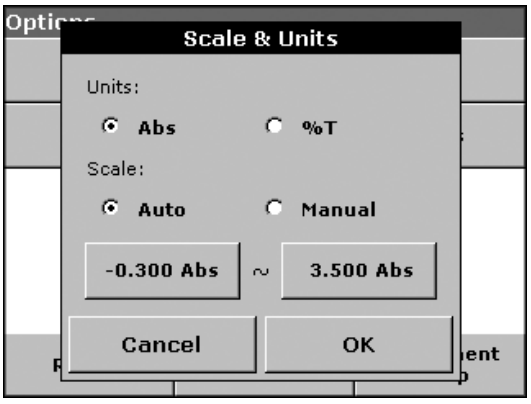
NOTE Do not enter the same value for lower and upper limit. When entering a value for the lower limit that is higher than the upper limit or vice versa, the values are reversed.

4. Select the scan resolution or step by touching the respective radio button. Selecting a larger step allows the instrument to scan faster, but decreases the resolution of the scan. Depending on the selected wavelength range, you may select steps between 0.1 and 5 nm.

NOTE A total of 910 data locations are available for each scan (1100 nm - 190 nm = 910) at a 1 nm step. Intervals that exceed this boundary are automatically grayed out and cannot be selected.

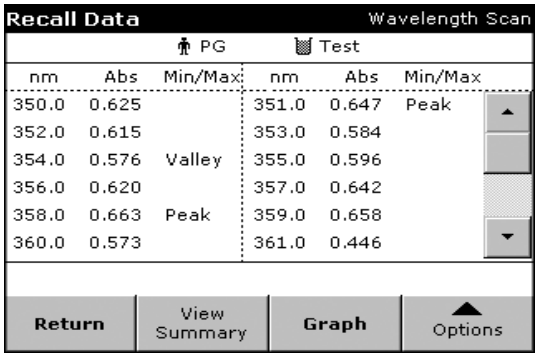
NOTE Recordings of high resolution scans take longer than recordings of low resolution scans. For example, 0.1 nm resolution is equal to a speed of 100 nm/min while 5 nm resolution is equal to a speed of 4500 nm/min. A common and practical scan interval or resolution is 1 nm, which is equal to a scan speed of 910 nm/min.

5. Touch **OK** to return to the scan mode.



To set scale and units for the y-axis:

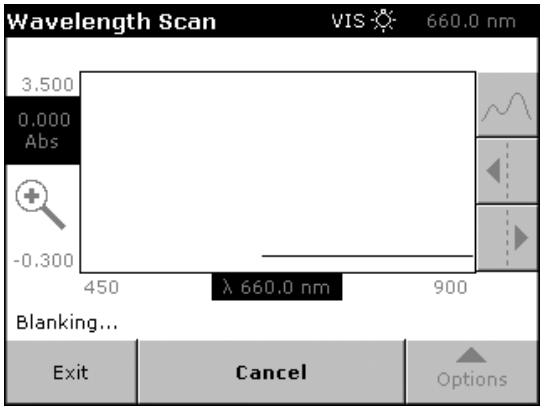
1. Touch **Options**, **More...**, and then **Scale & Units**.
2. Select **Abs** or **%T** by touching the radio button.
3. Select **Auto** or **Manual** scaling.
   - With **Auto** scaling (default), the screen scales the scan to fit the graph area.
   - With **Manual** scaling, you can set the lower and upper limits of the y-axis by touching the respective button and entering the appropriate value using the numeric keypad.
4. Touch **OK** to return to the Options screen.
5. Touch **Return** to return to the scan mode.



To toggle between the scan display (graph) and the tabulated scan data, touch **Select Table** or **Select Graph** in the Options menu after taking a scan.

6.2 Scanning

Before you can take a wavelength scan, you must first scan the baseline (performing a blank scan). Changing any of the scanning parameters requires a new baseline scan. After scanning the baseline, the instrument is ready to take sample scans.

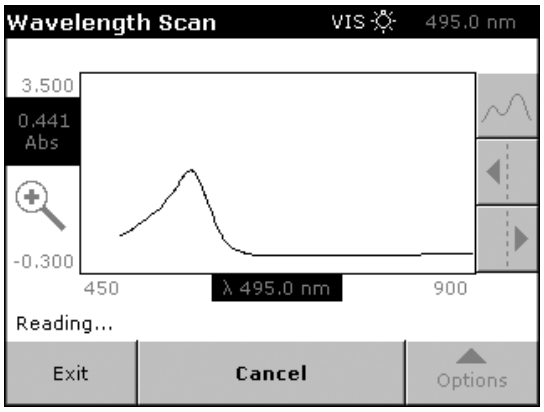


To enter the Wavelength Scan mode:

1. Touch **Wavelength Scan** in the Main Menu.
2. Insert the cuvette with a "blank" solution into the cell holder and close the cell compartment (recommended). To blank against air, use an empty cuvette or leave the cell holder empty.

NOTE If the lid of the cell compartment is open, the instrument disables the **Blank**/**Read** buttons.

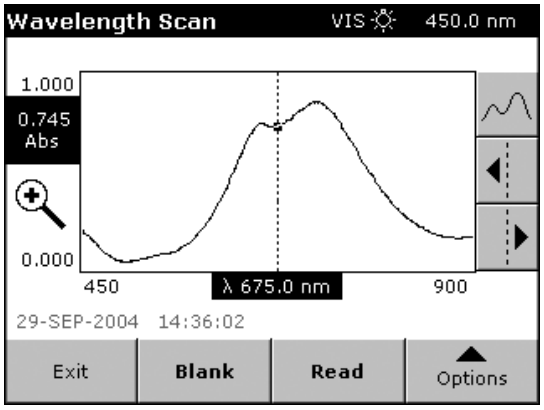
3. Touch **Blank** to start the baseline scan. The screen displays the "Blanking..." message in the bottom-left corner of the screen.
4. When the system has been blanked, insert a sample cuvette into the cell holder and close the cell compartment.
5. Touch **Read** to start the scan. The screen displays the "Reading..." message in the bottom-left corner of the screen.



NOTE During the scanning process, the **Read**/**Blank** buttons are replaced by a **Cancel** button. You may touch **Cancel** to stop and discard the scan in progress.

When the scan is complete, the instrument does the following:

1. Displays the entire scan over the selected wavelength range.
2. Scales the y-axis depending on the selected scaling mode.
3. Activates the cursor functions in the vertical navigation bar.



Located on the right side of the scan graph, three buttons allow you to navigate the scan:

- **Peak/Valley**: Touch the top button to toggle between the **Track** and **Peak/Valley** (button that includes arrows) cursor modes.

NOTE You can select the default cursor mode in the Options screen.

- **Left/Right arrow buttons**: The two arrow buttons in the vertical menu move the cursor left and right to the next or previous data point (**Track**) or the next or previous peak or valley (**Peak/Valley** mode).

The wavelength at the cursor position is highlighted at the x-axis of the graph and the corresponding Abs or %T is highlighted on the y-axis.

NOTE You can also move the cursor by directly touching on the displayed graph.

To zoom in (+) or out (-), touch the Zoom Symbol on the left side of the screen. Zoom enlarges the area around the cursor position. Touch the Zoom Symbol again to return to the former scale.

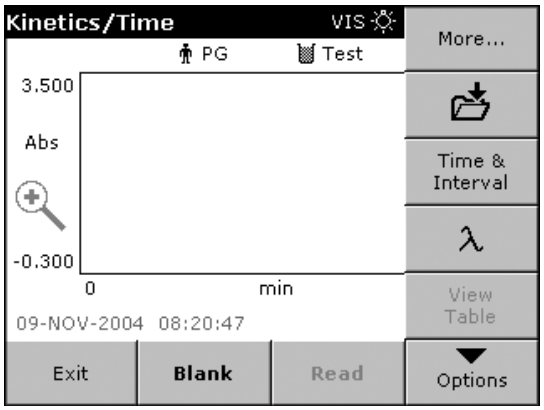
NOTE For instructions on using a Carousel Cell Holder, a Sipper, or a Peltier Temperature Controller, see "Carousel, Sipper, and Peltier Operation" on page 117.

NOTE For instructions on printing and data storage, see "Store, Recall, Send, and Delete Data" on page 125.

Kinetic/Time Mode

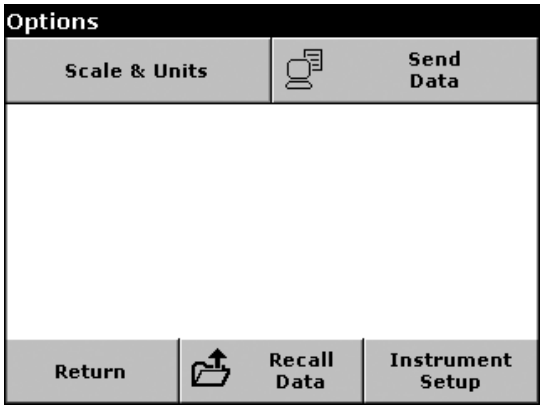
The Kinetic/Time mode lets you collect data in either absorbance or transmittance for a user-defined length of time. After collecting the data, you can display the kinetic profile or the tabular data and calculate the rate or slope of a given time range.

7.1 Parameter Setup



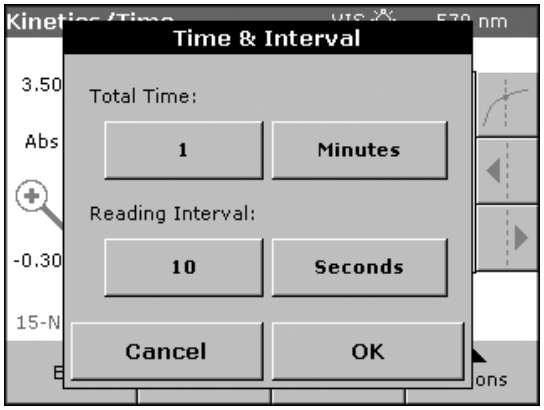
To enter the Kinetics/Time mode:

1. Touch **Kinetics/Time** mode in the Main Menu.
2. Touch **Options** to set the parameters.
   - **More...:** displays further options in the Options screen.
   - **Store icon:** stores the kinetic data.
   - **Time & Interval:** lets you define the total time for data collection and the time interval between the collected data points.
   - **λ:** lets you determine the wavelength.
   - **View Table / View Graph**: displays the tabular data or the graph. By default, the graph or kinetic profile appears. The button becomes enabled after the run is complete.



3. Touch **More...** to go to the Options screen.
   - **Scale & Units**: lets you select **Abs** or **%T** and determine the scaling of the graph (manual or automatic).
   - **Send Data**: sends data to a printer, computer, and storage device.

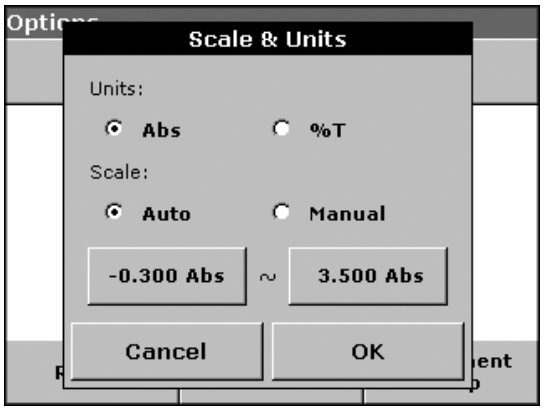
NOTE For instructions on storing and printing scan data, see "Store, Recall, Send, and Delete Data" on page 125.



To set the run time and interval:

1. Touch **Time & Interval** in the Options screen.
2. Touch the left buttons and enter the **Total Time** and the **Reading Interval** using the numeric keyboard.
3. Touch the right buttons to choose either **Minutes** or **Seconds**.
4. Touch **OK** to confirm the kinetics setup.

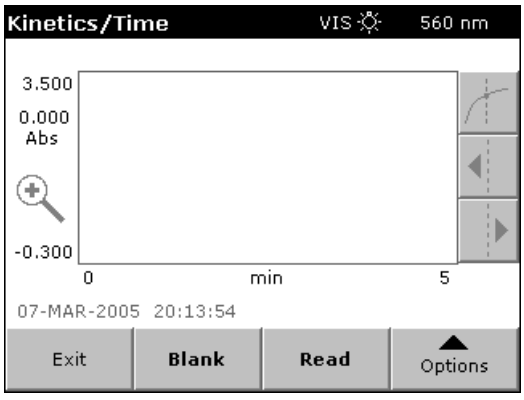
NOTE The Reading Interval must be 10 Seconds or larger. The kinetics run can have a maximum of 910 data points.



To set the scale and unit for the y-axis:

1. Touch **Options**, **More...**, and then **Scale & Units**.
2. Select **Abs** or **%T** by touching the radio button.
3. Select **Auto** or **Manual** scaling.
   - With **Auto** scaling (default), the screen scales the scan to fit the graph area.
   - With **Manual** scaling, you can set the lower and upper limit of the y-axis by touching the buttons at the bottom and entering the appropriate values using the numeric keypad.
4. Touch **OK** to return to the Options screen.
5. Touch **Return** to return to the measurement mode.

7.2 Taking the Kinetics Readings



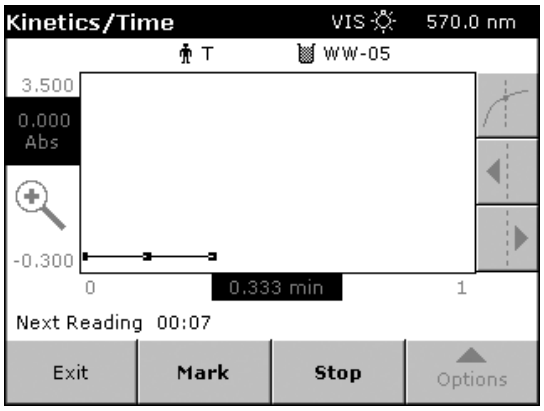
Before starting a kinetics run, you must blank the instrument. After blanking, the instrument is ready for the kinetics run.

To enter Kinetics/Time mode:

1. Touch **Kinetics/Time** mode in the Main Menu.
2. Insert a cuvette with a "blank" solution into the cell holder and close the cell compartment (recommended). To blank against air, use an empty cuvette or leave the cell holder empty.

NOTE If the lid of the cell compartment is open, the instrument disables the **Blank/Read** buttons.

3. Touch **Blank** to blank the instrument. The screen displays the "Blanking..." message in the bottom-left corner of the screen.



4. When the system has been blanked, insert the sample cuvette into the cell holder and close the cell compartment.

5. Touch **Read** to start the kinetics run. The screen displays the "Reading..." message in the bottom left corner of the screen with the count-down for the remaining time before the next data point is taken.

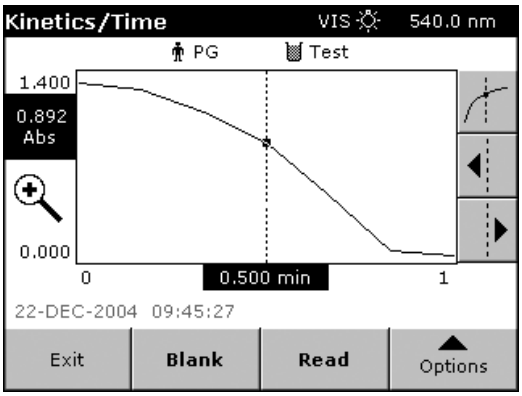
NOTE During the reading process, the **Read/Blank** buttons are replaced by a **Mark** and a **Stop** button. You may touch **Stop** to stop and discard the run in progress

- **Mark**: lets you mark the next data point collected. The instrument does not use this mark; it is provided so you can indicate a significant event, such as the addition of a sample or other reagent. The mark is also shown in the table.
- **Stop**: ends the kinetic run and discards the data.

7.3 Analysis of a Kinetic Run

When the kinetic run completes, the instrument does the following:

- Activates the sound, emitting an acoustic signal to let you know that the run is complete.
- Displays the graph over the entire time (total time).
- Scales the y-axis depending on the selected scaling mode.
- Activates the cursor functions in the vertical navigation bar.



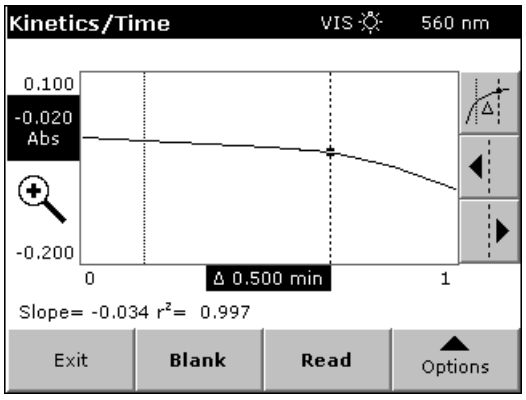
After completing a Kinetics/Time run, the instrument displays a graph of absorbance versus time. The graph shows one cursor, which is the Elapsed (t) (elapsed time) cursor mode.

Located at the right side of the kinetic profile, three buttons allow you to navigate the kinetic data:

- **Cursor Mode**: Touch the top button to toggle between the **Elapsed** (one point) and the Delta (Δt) (second point) cursor mode.
- **Left/Right Arrows**: The two arrow buttons on the vertical menu move the cursor to the next or previous time point. The time at the cursor position is highlighted at the x-axis of the graph and the corresponding Abs is highlighted on the y-axis.

NOTE You can also move the cursor by directly touching on the displayed graph.

- **Zoom Symbol**: To zoom in (+) or out (-), touch the Zoom Symbol on the left side of the screen. Zoom enlarges the area around the cursor position. Touch the Zoom Symbol again to return to the former scale.



To determine the rate or slope, toggle to the Delta (Δt) cursor mode with the top button on the vertical menu. In this mode, you can use the previous cursor position to mark one fixed limit and use the second cursor to determine the other limit. These limits determine the time range used to calculate the rate or slope, displayed at the bottom-left corner of the screen, together with the corresponding r^2 value (correlation coefficient).

NOTE For instructions on using a Carousel Cell Holder, a Sipper, or a Peltier Temperature Controller, see "Carousel, Sipper, and Peltier Operation" on page 117.

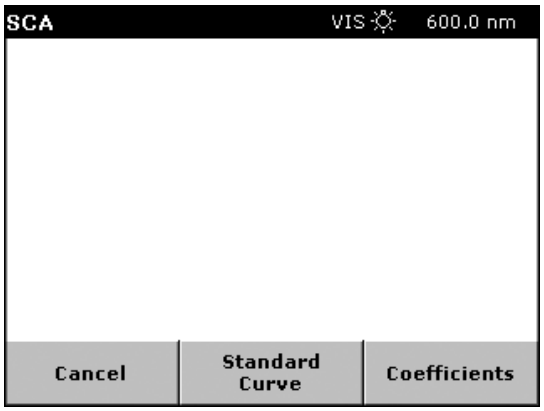
NOTE For instructions on printing and storing data, see "Store, Recall, Send, and Delete Data" on page 125.

Single Component Analysis Mode (SCA)

The Single Component Analysis (SCA) lets you calculate the concentrations from unknown samples containing a single component, using a standard curve. The standard curve can have up to 24 standards with known concentrations. The data from the standards fit to either a linear or nonlinear (quadratic) curve.

This mode requires a standard curve before analyzing the samples. You can specify the standard curve either by measuring standards with known concentrations or by entering the coefficients from a previous standard curve.

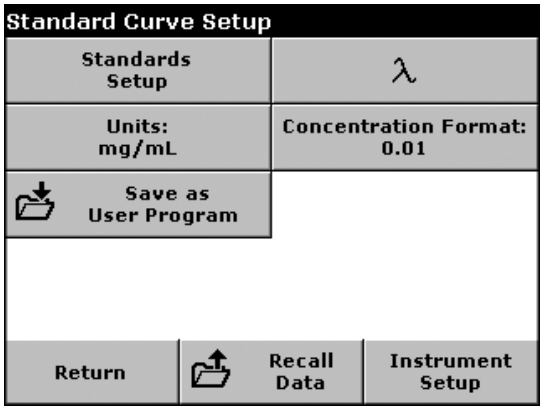
8.1 Parameter Setup



To enter the Single Component Analysis mode, touch **Single Component Analysis** in the Main Menu.

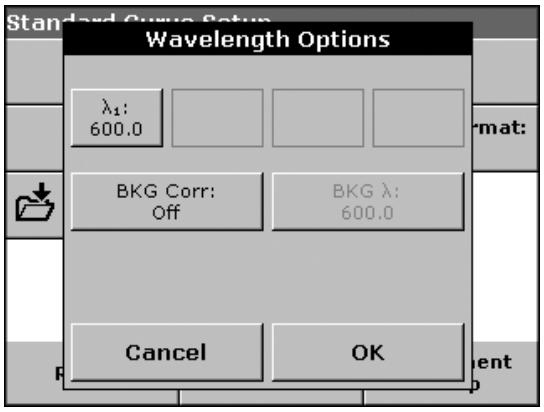
- Touch **Standard Curve** if you want to create a standard curve by measuring standards with known concentrations. (For details, see "Standard Curve Setup" on page 61.)
- Touch **Coefficients** if you want to enter the coefficients from a previous standard curve. (For details, see "Coefficients Setup" on page 64.)

Standard Curve Setup



Use the Standard Curve Setup screen to set the parameters.

- **Standards Setup:** allows you to enter the concentrations for each standard. This also determines the number of data points you will have for the standard curve.
- **λ:** lets you enter the wavelength and turn background correction off (default) or on.
- **Units:** allows you to select the desired concentration unit and/or define new units.
- **Concentration Format:** lets you determine the number significant digits for the concentration values.
- **Save as User Program:** stores the selected parameters as a user program (see "Store, Recall, Send, and Delete Data" on page 125).



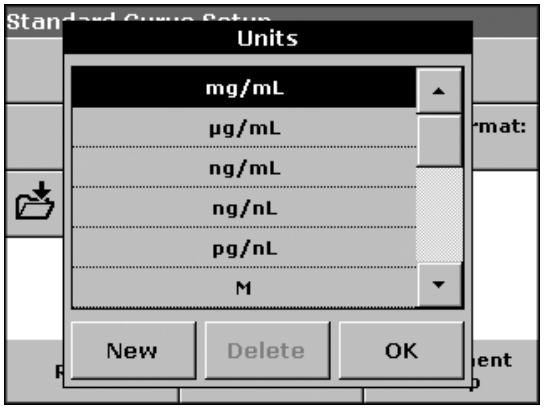
Touch λ, on the Standard Curve Setup screen to display the Wavelength Options screen.

- λ_1: displays the current wavelength. To change it, touch λ_1 and enter the desired wavelength using the numeric keypad.
- **BKG Corr:** toggles background correction between on and off. When active, you can set the background wavelength by touching **BKG** λ.

NOTE With background correction active, the analysis subtracts the absorbance reading at the background wavelength (Abs_{bkg}) from the absorbance reading at the analytical wavelength (Abs_{wl}).

The system reports the net absorbance readings (Abs_{net}) and uses them for all calculations:

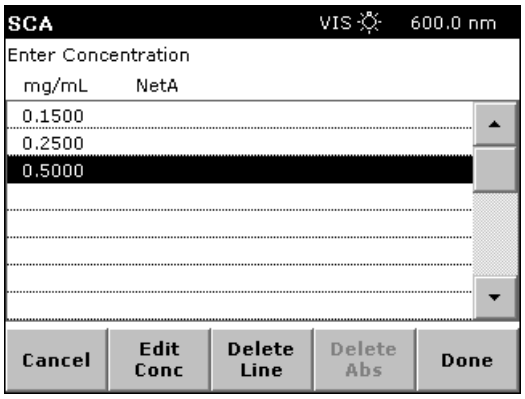
$$Abs_{net} = Abs_{wl} - Abs_{bkg}$$



To change the unit, touch **Units** in the Standard Curve Setup screen.

- To select an existing unit, touch the line (scroll down, if necessary).
- To enter a new unit, touch **New** and enter the unit using the alphanumeric keyboard.
- To delete a unit, touch its description to highlight it, then touch **Delete**.

NOTE The pre-defined units are fixed. You may delete only user-defined units.

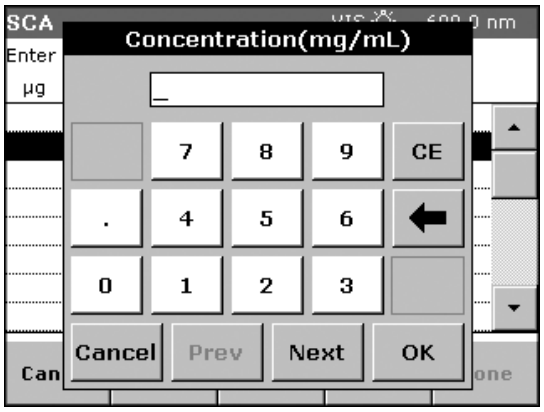


If you had selected **Standards Setup,** the SCA screen appears. Use this screen to enter the known concentration for each standard.

By default, the first line is highlighted. After you confirm a concentration entered with the numeric keypad, the screen highlights the next line automatically for the next entry.

NOTE You can highlight a desired line at any time by touching it if you need to edit or enter a concentration or modify a specific line.

- **Edit Conc:** lets you edit or enter the concentration using the numeric keypad.
- **Delete Line**: removes the selected line. The button is enabled only when a line with a concentration is highlighted.
- **Delete Abs**: this button remains disabled until standards have been read and absorbance values are available.



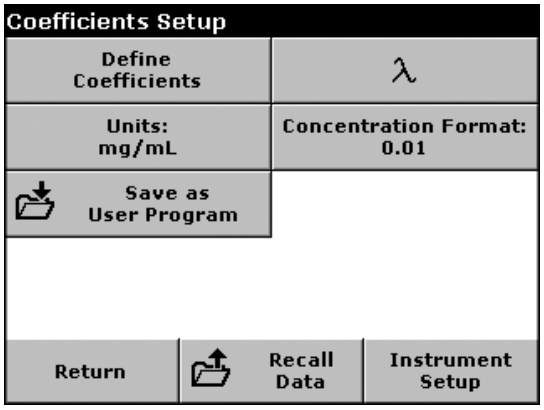
This numeric keypad appears when you touch **Edit Conc**.

Enter the desired concentration and touch **Next** to enter the next value. The system adds the concentration to the list (in the background) and highlights the next line. Continue until all concentrations have been entered.

- **Next**: steps forward in the entry list and displays the corresponding concentration, if available.
- **Prev**: steps back in the entry list and displays the corresponding concentration, if available.
- **Cancel**: discards the current entry and returns to the previous screen.

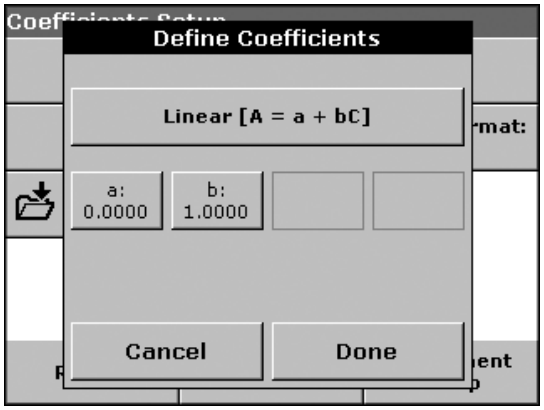
When you are done entering all concentrations (or making modifications), touch **OK** to return to the Enter Concentration list. Then touch **Done** to start reading the standard cuvettes and creating the standard curve.

Coefficients Setup



Touch **Coefficients** in the SCA screen to display the Coefficients Setup screen. You may choose the following options:

- **Define Coefficients:** allows you to enter the coefficients of a known standard curve.
- **λ:** lets you enter the wavelength and turn background correction off (default) or on.
- **Units:** allows you to select the desired concentration unit and/or define new units.
- **Concentration Format:** lets you determine the number of significant digits for the concentration values.
- **Save as User Program:** stores the selected parameters as a user program (see "Store, Recall, Send, and Delete Data" on page 125).

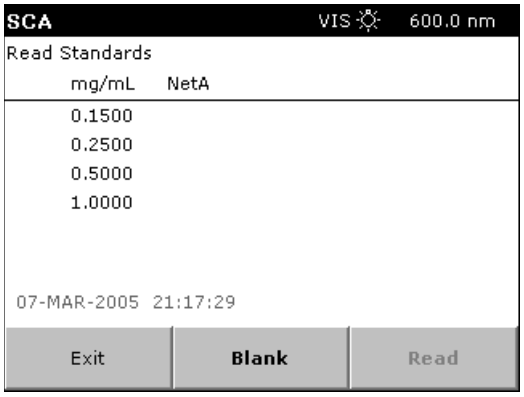


The Define Coefficients screen allows you to select the curve fit and enter the known coefficients [**a** and **b** for a linear fit and **a**, **b**, and **c** for a non-linear (quadratic) fit]. To use this screen:

1. Touch **Linear/Non-Linear** and select the desired curve fit (equation).
2. Touch each of the enabled buttons (**a:**, **b:**, **c:**) and enter the respective coefficient using the numeric keyboard.
3. Touch **Done** when the setup is complete.

A standard curve is now available and you can measure unknown samples and calculate the concentrations.

8.2 Reading Standards

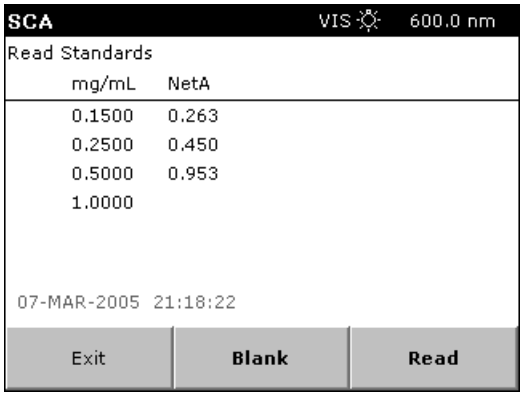


After you have entered the concentrations for the standards, the system is ready to read the standard cuvettes.

1. Insert a cuvette with a "blank" solution into the cell holder and close the cell compartment (recommended). To blank against air, use an empty cuvette or leave the cell holder empty.

NOTE If the lid of the cell compartment is open, the instrument disables the **Blank/Read** buttons.

2. Touch **Blank** to blank the instrument. The screen displays the "Blanking..." message in the bottom-left corner of the screen.



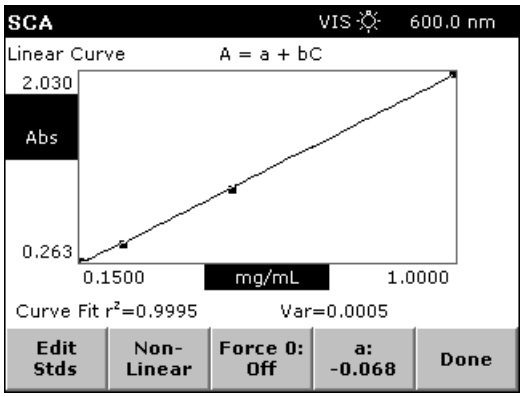
3. When the system has been blanked, insert the first standard cuvette into the cell holder and close the cell compartment.

4. Touch **Read** to start the reading. The screen displays the "Reading..." message to the right of the concentration and adds the reading (Net Abs) to the line with the respective concentration value.

NOTE Make sure you insert the standard cuvettes in the right sequence, based on the entered concentration.

NOTE During the reading process, the **Read/Blank** buttons are replaced by a **Cancel** button. You may touch **Cancel** to stop and discard the current reading.

5. Continue until all standard cuvettes have been read and an absorbance value is associated with each concentration.



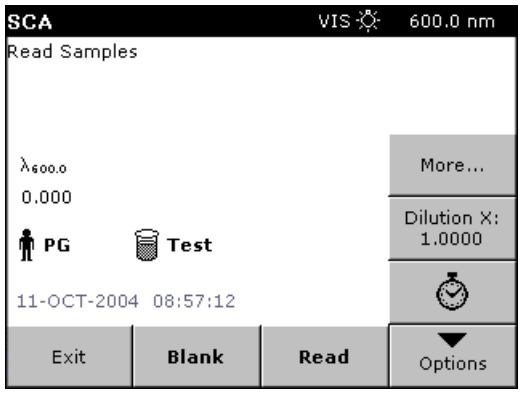
After reading the last standard, the system calculates and displays the standard curve.

- **Edit Stds**: goes to the Enter Concentrations screen. The list in this screen now has absorbance values for each concentration. You may edit information and/or re-read standards.
- **Non-Linear/Linear**: toggles between **Linear** and **Non-Linear** curve fit. By default, the system applies the **Linear** curve fit.
- **Force 0 (Off/On)**: allows you to force the curve fit through zero.
- **a:**, **b:**, **c:** displays the coefficients of the standard curve. The screen displays only one coefficient at a time. Touch the button to see the next coefficient.

The bottom of the screen displays the r^2 value (correlation coefficient) for this standard curve, together with the variance (**Var**). This allows you to judge the quality of the curve fit. Try different parameters (such as **Force 0** and/or **Linear** or **Non-Linear** curve fit) to see if the quality of the curve fit can be improved.

When you are satisfied with your standard curve, touch **Done** to store the coefficients and to start analyzing your samples.

8.3 Analyzing Samples



NOTE If the **Read** button is grayed out, you need to blank the system before you can read samples.

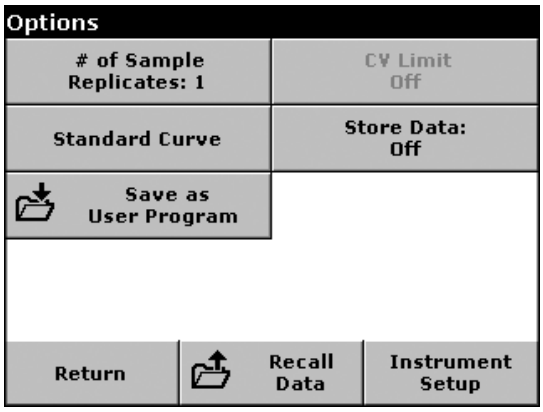
1. Insert a sample cuvette into the cell holder and close the cell compartment.
2. Touch **Read** to read the sample. The screen displays the result with the corresponding unit.

NOTE During the reading process, the **Read/Blank** buttons are replaced by a **Cancel** button. You may touch **Cancel** to stop and discard the current reading.

3. Touch **Options** to access parameters for the sample readings:

   - **More...**: provides more parameters in the Options screen.
   - **Dilution X**: lets you enter a dilution correction factor for the samples. The displayed results are corrected for dilution using the following equation:

     Con_{corr} = Conc x Dilution.
   - **Timer Icon**: sets the timer. Using the timer does not affect the readings that the instrument is taking. The times are not stored with sample readings.



4. Touch **More...** to go to the Options screen.

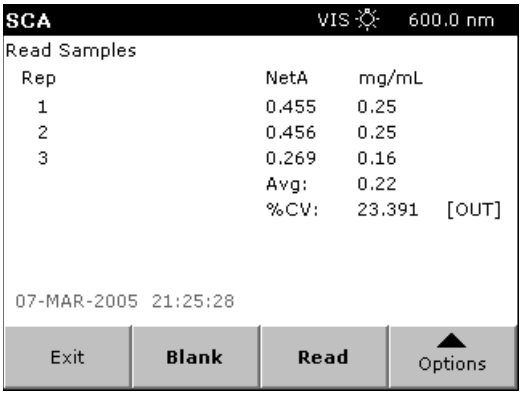
   - **# of Sample Replicates**: lets you enter the number of sample replicates (1 to 3).

NOTE When analyzing two replicates, the instrument calculates the average. When analyzing three replicates, the instrument calculates the mean and the coefficient of variation (CV), if enabled.

   - **CV Limit (Off/On)**: If three replicates are selected, the button CV Limits will be enabled and you can toggle the reporting of the Coefficient of Variation (CV) limit on or off and set a CV limit. Samples that are outside of the specified CV limit are flagged with [OUT].
   - **Standard Curve**: allows you to the Standard Curve Setup parameters.

NOTE You can only view the standard curve; you cannot edit it.

   - **Store Data**: stores the sample data (see "Store, Recall, Send, and Delete Data" on page 125).
   - **Save as User Program**: stores the selected parameters as a user program (see "User Programs" on page 43).



This screen shows the results of one sample with three replicates. The CV Limit option has been turned on and the sample is flagged because the value is outside the accepted limit.

NOTE For using a Carousel Cell Holder, a Sipper, or a Peltier Temperature Controller, see "Carousel, Sipper, and Peltier Operation" on page 117.

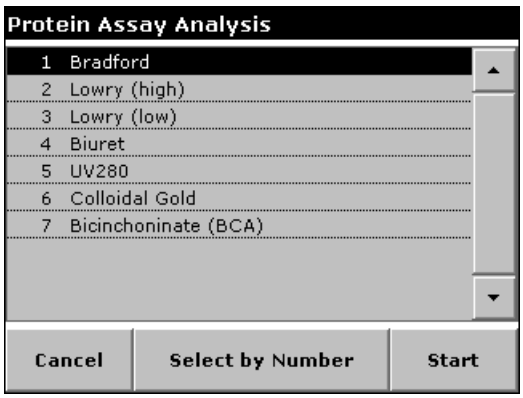
NOTE For printing and storing data, see "Store, Recall, Send, and Delete Data" on page 125.

Protein Assay Analysis

The Protein Assay Analysis mode provides pre-programmed methods for common protein assays to calculate the amount of protein in a sample. The standard curve can have up to 24 standards with known concentrations. The data from the standards fit to either a linear or nonlinear (quadratic) curve.

This mode requires a standard curve before samples can be analyzed. You can create the standard curve either by measuring standards with known concentrations or by entering the coefficients from a previous standard curve.

9.1 Method Description



Protein Assay Analysis provides methods described in the following sections:

- "Bradford" on page 69
- "Lowry (High)" on page 69
- "Lowry (Low)" on page 70
- "Biuret" on page 70
- "UV280" on page 70
- "Colloidal Gold" on page 70
- "Bicinchoninate (BCA)" on page 70

Bradford

The Bradford method for measuring total protein concentration is based on a colorimetric reaction between Coomassie Brilliant Blue G-250 and the protein in the sample. The complex is formed in about 2 minutes and is stable for 1 hour. A wavelength of 595 nm is used. From 1 to 140 mg/ml of protein can be measured. There are few interferences. This analysis is about four times more sensitive than the Lowry analysis.

Lowry (High)

The Lowry method for measuring protein concentration is a colorimetric procedure. Protein is first reacted with an alkaline copper reagent, then with the Folin phenol reagent at room temperature. The blue copper complex is read at 750 nm for samples containing less than 25 mg/ml protein. This analysis is 10 to 20 times more sensitive than measurement at 280 nm, and 100 times more sensitive than the Biuret analysis.

The following substances react with the Folin reagent: tryptophan, tyrosine, most phenols, uric acid, guanine, and xanthine.

Lowry (Low)

This method is the same as the Lowry discussed above, except that the samples containing more than 25 g/ml protein are analyzed at 500 nm.

Biuret

The Biuret method forms a blue copper complex under alkaline conditions. From 10 to 1,200 g/ml can be measured at 540 nm. The Biuret method is less sensitive to amino acid composition than the Lowry method. There are practically no biological substances with significant interference at pH 7.1.

UV280

Virtually all proteins have an absorption maxima at neutral pH around 280 nm due to absorption by tyrosine (λ_{min} = 275 nm), phenylalanine (lambda max = 257 nm), and tryptophan (λ_{max} = 279 nm). Since proteins differ in their content of these amino acids, this analysis is only approximate, unless the standards used to construct the standard curve are made using the same protein measured in the samples. This assay is linear in the range of 50 to 3,000 g/ml.

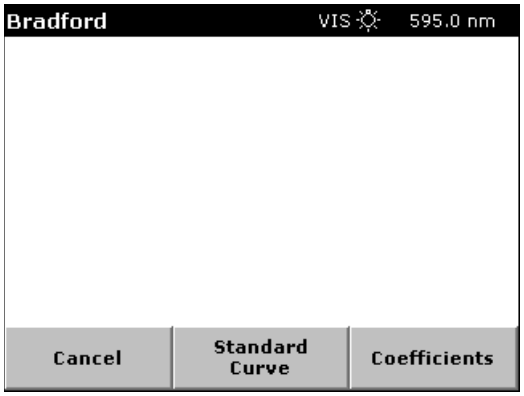
Colloidal Gold

Proteins form a stable complex with colloidal gold, which allows very small quantities of protein to be analyzed at 595 nm. This assay is linear in the range of 5 to 200 ng/ml.

Bicinchoninate (BCA)

The BCA assay involves the formation of the Biuret protein-copper complex in alkaline solution. This complex is then reacted with bicinchoninic acid (BCA) to form a water soluble salt, that has a strong absorbance at 562 nm. This assay is easier to perform than the Lowry assay. The standard assay has an applicable protein concentration range of 20 to 1,200 g/ml. The micro assay has an applicable protein concentration range of 0.5 to 20 g/ml. When working in the full range, a non-linear standard curve calculation is suggested.

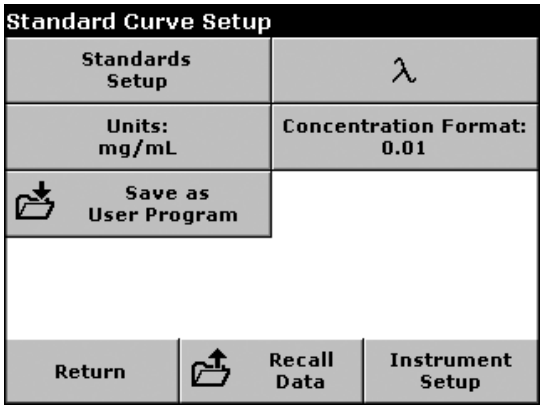
9.2 Parameter Setup



To enter the Protein Assay Analysis mode, touch **Protein Assay Analysis** in the Main Menu.

- Touch **Standard Curve** if you want to create a standard curve by measuring standards with known concentrations. (For details, see "Standard Curve Setup" below.)
- Touch **Coefficients** if you want to enter the coefficients from a previous standard curve. (For details, see "Coefficients Setup" on page 74.)

Standard Curve Setup

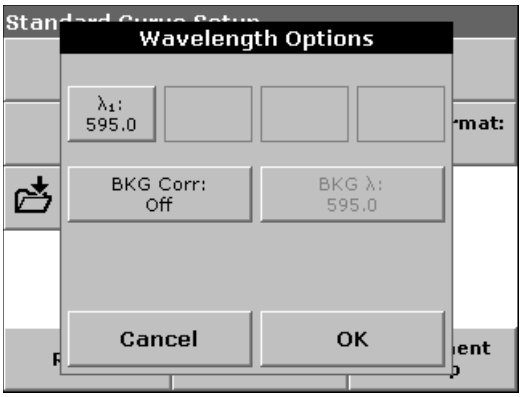


Use the Standard Curve Setup screen to set the parameters.

- **Standards Setup:** allows you to enter the concentrations for each standard. This also determines the number of data points you will have for the standard curve.
- **λ:** lets you enter the wavelength and turn background correction off (default) or on.

NOTE The appropriate wavelength is set automatically for the selected assay. It is recommended not to change this default wavelength unless you are running an assay that is not available as a method.

- **Units:** allows you to select the desired concentration unit and/or define new units.
- **Concentration Format:** lets you determine the number significant digits for the concentration values.
- **Save as User Program:** stores the selected parameters as a user program (see "Store, Recall, Send, and Delete Data" on page 125).



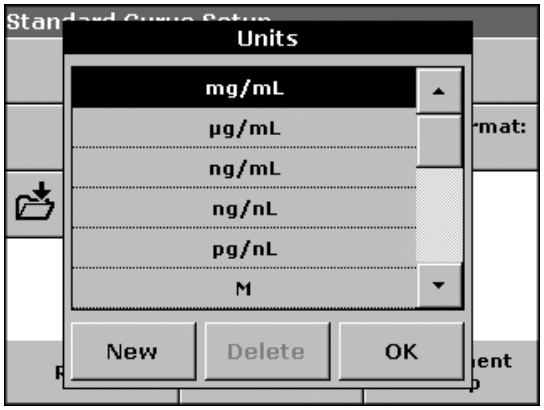
Touch λ_1 on the Standard Curve Setup screen to display the Wavelength Options screen.

- λ_1: displays the current wavelength. To change it, touch λ_1 and enter the desired wavelength using the numeric keypad.
- **BKG Corr**: toggles background correction between on and off. When active, you can set the background wavelength by touching the **BKG** λ button.

NOTE With background correction active, the analysis subtracts the absorbance reading at the background wavelength (Abs_{bkg}) from the absorbance reading at the analytical wavelength (Ab_{swl}).

The system reports the net absorbance readings (Abs_{net}) and uses them for all calculations:

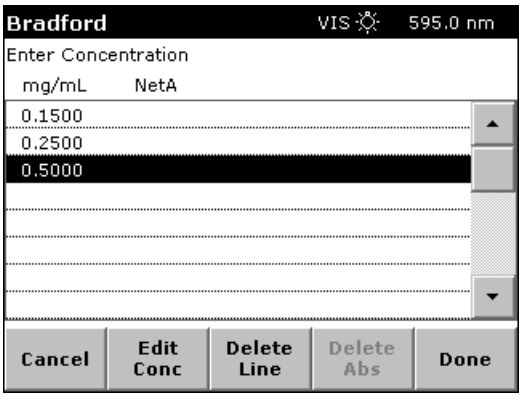
$$Abs_{net} = Ab_{swl} - Abs_{bkg}$$



To change the unit, touch **Units** in the Standard Curve Setup screen.

- To select an existing unit, touch the line (scroll down, if necessary).
- To enter a new unit, touch **New** and enter the unit using the alphanumeric keyboard.
- To delete a unit, touch its description to highlight it, then touch **Delete**.

NOTE The pre-defined units are fixed. You may delete only user-defined units.

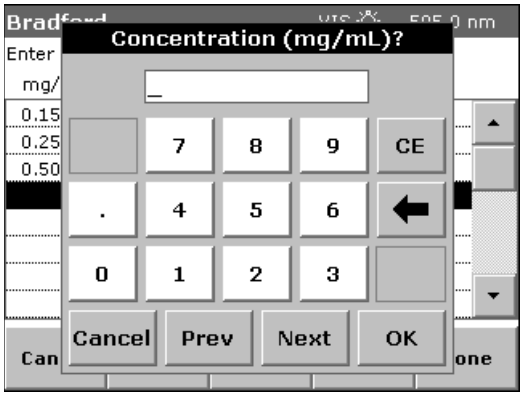


If you had selected **Standards Setup**, a screen with the selected method appears. Use this screen to enter the known concentration for each standard.

By default, the first line is highlighted. After you confirm a concentration entered with the numeric keypad, the screen highlights the next line automatically for the next entry.

NOTE You can highlight a desired line at any time by touching it if you need to edit or enter a concentration or modify a specific line.

- **Edit Conc**: lets you edit or enter the concentration using the numeric keypad.
- **Delete Line**: removes the selected line. The button is enabled only when a line with a concentration is highlighted.
- **Delete Abs**: this button remains disabled until standards have been read and absorbance values are available.



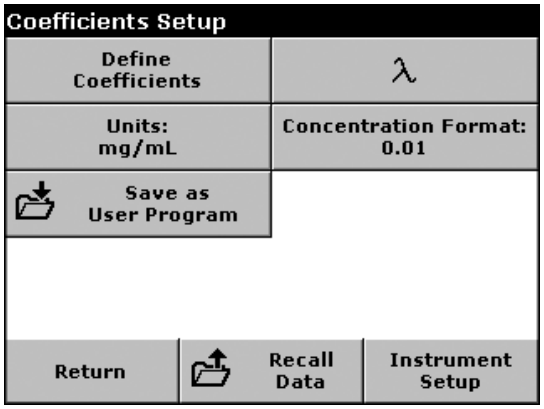
This numeric keypad appears when you touch **Edit Conc**.

Enter the desired concentration and touch **Next** to enter the next value. The system adds the concentration to the list (in the background) and highlights the next line. Continue until all concentrations have been entered.

- **Next**: steps forward in the entry list and displays the corresponding concentration, if available.
- **Prev**: steps back in the entry list and displays the corresponding concentration, if available.
- **Cancel**: discards the current entry and returns to the previous screen.

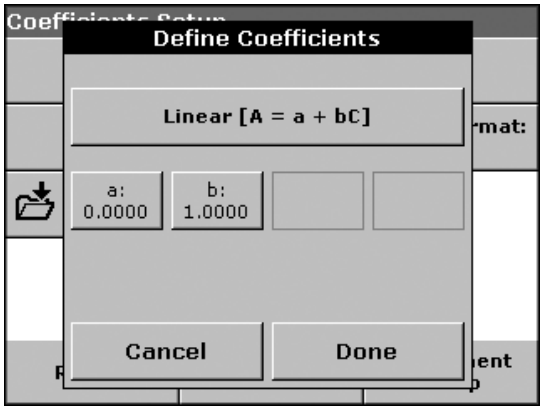
When you are done entering all concentrations (or making modifications), touch **OK** to return to the Enter Concentration list. Then touch **Done** to start reading the standard cuvettes and creating the standard curve.

Coefficients Setup



Touch **Coefficients** in the Protein Assay Analysis screen to display the Coefficients Setup screen. You may choose the following options:

- **Define Coefficients:** allows you to enter the coefficients of a known standard curve.
- **λ:** lets you enter the wavelength and turn background correction off (default) or on.
- **Units:** allows you to select the desired concentration unit and/or define new units.
- **Concentration Format:** lets you determine the number of significant digits for the concentration values.
- **Save as User Program:** stores the selected parameters as a user program (see "Store, Recall, Send, and Delete Data" on page 125).

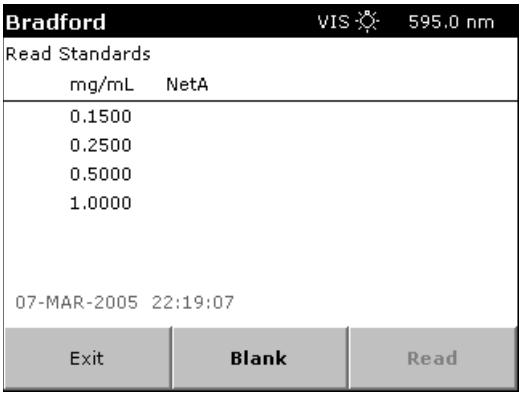


The Define Coefficients screen allows you to select the curve fit and enter the known coefficients [**a** and **b** for a linear fit and **a**, **b**, and **c** for a non-linear (quadratic) fit]. To use this screen:

1. Touch **Linear/Non-Linear** and select the desired curve fit (equation).
2. Touch each of the enabled buttons (**a:**, **b:**, **c:**) and enter the respective coefficient using the numeric keyboard.
3. Touch **Done** when the setup is complete.

A standard curve is now available and you can measure unknown samples and calculate the concentrations.

9.3 Reading Standards

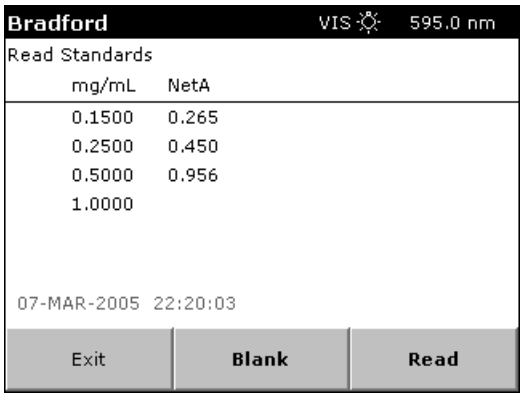


After you have entered the concentrations for the standards, the system is ready to read the standard cuvettes.

1. Insert a cuvette with a "blank" solution into the cell holder and close the cell compartment (recommended). To blank against air, use an empty cuvette or leave the cell holder empty.

NOTE If the lid of the cell compartment is open, the instrument disables the **Blank/Read** buttons.

2. Touch **Blank** to blank the instrument. The screen displays the "Blanking..." message in the bottom-left corner of the screen.



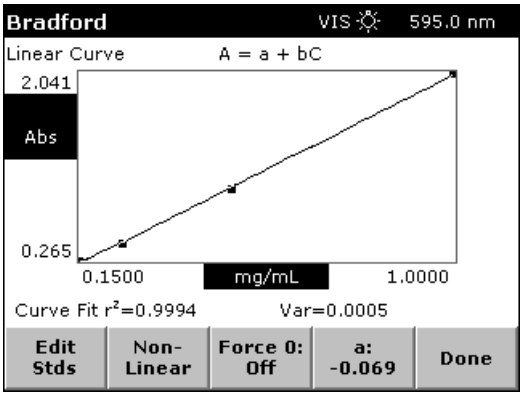
3. When the system has been blanked, insert the first standard cuvette into the cell holder and close the cell compartment.

4. Touch **Read** to start the reading. The screen displays the "Reading..." message to the right of the concentration and adds the reading (Net Abs) to the line with the respective concentration value.

NOTE Make sure you insert the standard cuvettes in the right sequence, based on the entered concentration.

NOTE During the reading process, the **Read/Blank** buttons are replaced by a **Cancel** button. You may touch **Cancel** to stop and discard the current reading.

5. Continue until all standard cuvettes have been read and an absorbance value is associated with each concentration.



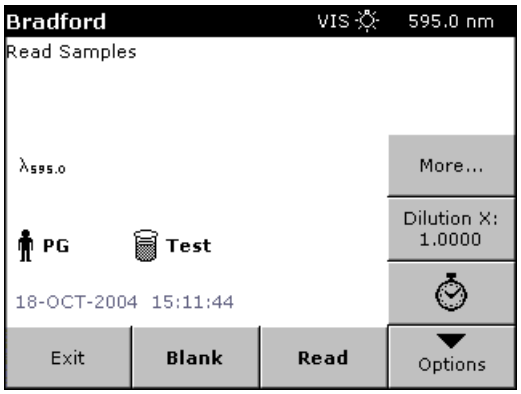
After reading the last standard, the system calculates and displays the standard curve.

- **Edit Stds**: goes to the Enter Concentrations screen. The list in this screen now has absorbance values for each concentration. You may edit information and/or re-read standards.
- **Non-Linear/Linear**: Toggles between **Linear** and **Non-Linear** curve fit. By default, the system applies the **Linear** curve fit.
- **Force 0 (Off/On)**: allows you to force the curve fit through zero.
- **a:**, **b:**, **c:** displays the coefficients of the standard curve. The screen displays only one coefficient at a time. Touch the button to see the next coefficient.

The bottom of the screen displays the r^2 value (correlation coefficient) for this standard curve, together with the variance (**Var**). This allows you to judge the quality of the curve fit. Try different parameters (such as **Force 0** and/or **Linear** or **Non-Linear** curve fit) to see if the quality of the curve fit can be improved.

When you are satisfied with your standard curve, touch **Done** to store the coefficients and to start analyzing your samples.

9.4 Analyzing Samples



NOTE If the **Read** button is grayed out, you need to blank the system before you can read samples.

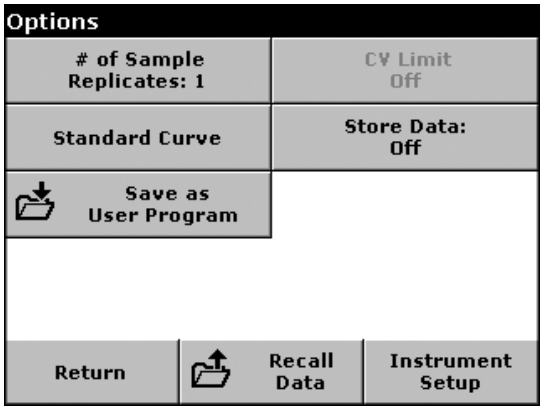
1. Insert a sample cuvette into the cell holder and close the cell compartment.
2. Touch **Read** to read the sample. The screen displays the result with the corresponding unit.

NOTE During the reading process, the **Read/Blank** buttons are replaced by a **Cancel** button. You may touch **Cancel** to stop and discard the current reading.

3. Touch **Options** to access parameters for the sample readings.

   - **More...**: provides more parameters in the Options screen.
   - **Dilution X**: lets you enter a dilution correction factor for the samples. The displayed results are corrected for dilution using the following equation:

     Con_{corr} = Conc x Dilution.
   - **Timer Icon**: sets the timer. Using the timer does not affect the readings that the instrument is taking. The times are not stored with sample readings.



4. Touch **More...** to go to the Options screen.

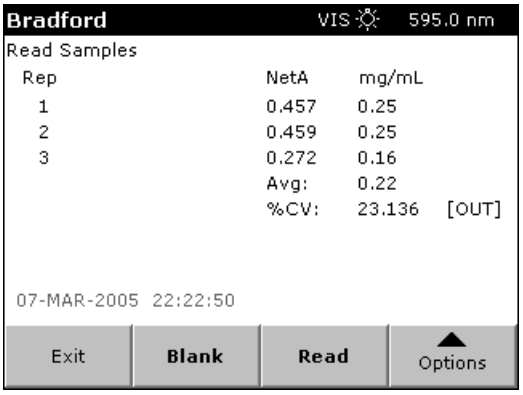
- **# of Sample Replicates**: lets you enter the number of sample replicates (1 to 3).

NOTE When analyzing two replicates, the instrument calculates the average. When analyzing three replicates, the instrument calculates the mean and the coefficient of variation (CV), if enabled.

- **CV Limit (Off/On)**: If you select three replicates, the **CV Limits** button becomes enabled. Use this button to toggle Coefficient of Variation (CV) limit reporting on or off and to set a CV limit. The sample result will flag samples outside of the specified CV limit with [OUT].
- **Standard Curve**: allows you to the Standard Curve Setup parameters.

NOTE You can only view the standard curve; you cannot edit it.

- **Store Data**: stores the sample data (see "Store, Recall, Send, and Delete Data" on page 125).
- **Save as User Program**: stores the selected parameters as a user program (see "User Programs" on page 43).



This screen shows the results of one sample with three replicates. For this example, the **CV Limit** option has been turned on; because the value is outside the CV limit, its value is flagged [OUT].

NOTE For using a Carousel Cell Holder, a Sipper, or a Peltier Temperature Controller, see "Carousel, Sipper, and Peltier Operation" on page 117.

NOTE For printing and storing data, see "Store, Recall, Send, and Delete Data" on page 125.

Nucleic Acid Analysis

The Nucleic Acid Analysis mode provides methods for DNA, RNA, and oligonucleotide analysis, including the calculation of absorbance ratios and concentrations. Some of the methods also determine molecular weight, nucleotide length, extinction coefficient, concentration, and theoretical melting temperature.

10.1 Method Description

NOTE When calculating a concentration from λ_1 (default is 260 nm), the wavelength should not be changed, according to text books and articles on the subject.

NOTE All methods in this mode require the UV lamp. You may switch it on manually and let it warm-up sufficiently for best results. If not turned on, the instrument turns the lamp on automatically before taking the blank measurement, using the minimum warm-up time. The UV Lamp icon appears in the display.

260/280 Ratio

The 260/280 Ratio method allows you to calculate a 260/280 absorbance ratio with optional background correction at 320 nm.

Single Ratio

The Single Ratio method allows you to calculate an absorbance ratio using any two wavelengths of choice, with an optional background wavelength.

Double Ratio & Concentration

The Double Ratio & Concentration method allows you to specify three analytical wavelengths and an optional background wavelength.

The results include:

- Absorbance ratio of λ_1/λ_2
- Absorbance ratio of λ_1/λ_3
- Concentration (λ_1 of 260) = λ_1 x factor

dsDNA, ssDNA, and RNA

These three methods allow you to calculate a 260/280 absorbance ratio with the optional use of background correction at 320 nm. You can change each of these wavelengths. The concentration is estimated in units of g/mL by multiplying the absorbance of the analytical wavelength by a factor.

The default factor is 50.0 for the dsDNA method, 33.0 for the ssDNA method and 40.0 for the RNA method. It is suggested that the default factor be used. However, you can change the factor.

Oligo DNA Long and Oligo RNA Long

These two methods allow you to calculate the purity, absorptivity (extinction coefficient), concentration and melting temperature (DNA only), based on the base composition and the absorbance at 260 nm. For equations, see "Calculations" on page 91.

These methods use 260 and 280 nm as the analytical wavelengths and 320 nm as the optional wavelength for background correction. You may change the second analytical wavelength (λ_2) and the background wavelength. The analytical wavelength of 260 nm is used to determine the concentration and cannot be changed.

NOTE "Long" oligonucleotides are considered those with a length of more than 30 bases. In this method, the calculation of the Theoretical Melting Point (T_m) for DNA can be far from the "true" value when the oligo is very short.

Oligo DNA Short and Oligo RNA Short

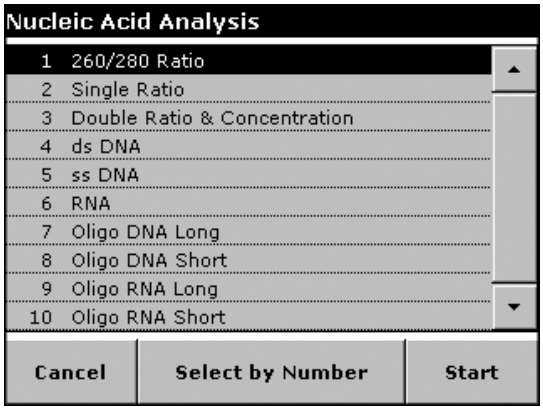
These two methods allow you to calculate the purity, absorptivity (extinction coefficient), concentration, and melting temperature (DNA only), based on the base sequence and the absorbance at 260 nm. Short nucleotides are typically between 10 and 30 bases. The maximum allowable value is 40 bases. For equations, see "Calculations" on page 91.

The methods use 260 and 280 nm analytical wavelengths and 320 nm as the optional wavelength for background correction. You can change the second analytical wavelength ((λ_2) and the background wavelength. The analytical wavelength of 260 nm is used to determine the concentration and cannot be changed.

NOTE "Short" oligonucleotides are considered those with a length of less than 30 bases. The Oligo DNA Short and Oligo RNA Short methods apply the nearest-neighbor* algorithm for best results.

* (DNA) Breslauer, K.J.; Frank, R.; Blöcker, H.; Marky, L.A. Proc. Natl. Acad. Sci. USA 83, 3746-3750(1986)
(RNA) Freier, S.M.; Kierzek, R.; Jaeger, J.A.; Sugimoto, N.; Caruthers, M.H.; Neilson, T.; Turner, D.H. Proc. Natl. Acad. Sci. 83, 9373-9377 (1986).

10.2 Parameter Setup

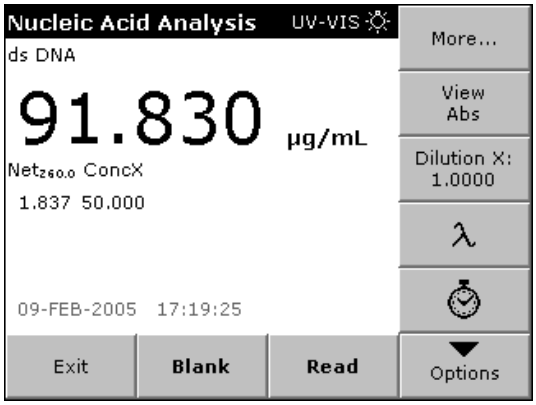


To enter the Nucleic Acid Analysis mode:

1. Touch **Nucleic Acid Analysis** in the Main Menu.
2. Touch the line with the method of choice to highlight it or touch **Select by Number** and enter the desired number.
3. Touch **Start** to enter the selected method.

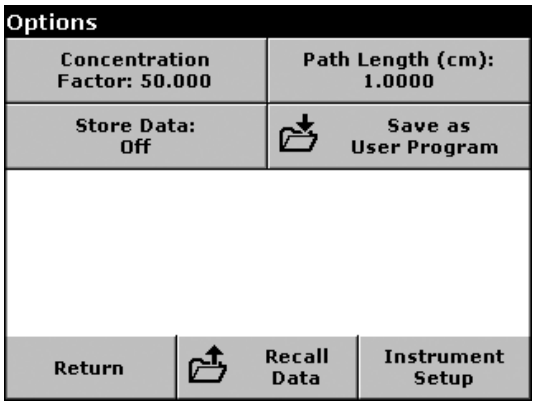
NOTE The name of the selected method appears on the top-left side of the display in the following screens.

260/280 Ratio and Single Ratio



Touch **Options** for the parameter setup.

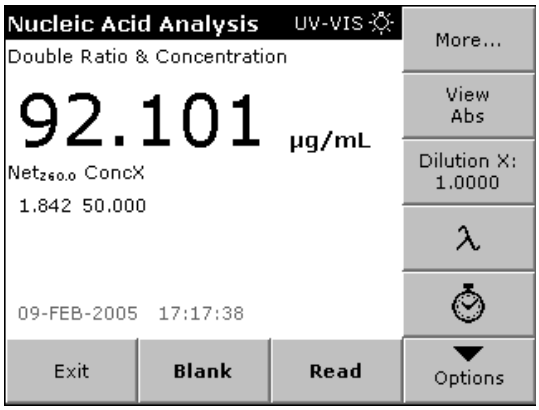
- **More...**: displays further options in the Options screen.
- For 260/280 Ratio, **BKG Corr (Off/On** lets you toggle background correction on and off. The wavelengths are fixed.
- For Single Ratio, λ lets you modify the default wavelengths and toggle background correction on and off. When background correction is on, you can modify the default background wavelength.
- **Timer Icon**: sets the timer. Using the timer does not affect the readings that the instrument is taking. The times are not stored with sample readings.



Touch **More...**. to go to the Options screen.

- **Path Length (cm):** allows you to modify the default path length of 1 cm if you want to use small path length cells for very low concentrations.
- **Store Data (Off/On)**: lets you toggle the storage setting and manually store data.
- **Save as User Program**: stores the selected parameters as a user program (see "User Programs" on page 43).

Double Ratio & Concentration



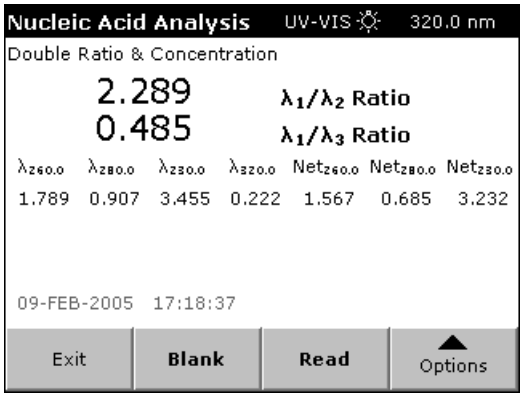
Touch **Options** for parameter setup.

- **More...**: displays further options in the Options screen.
- **View Conc/Abs**: lets you toggle between the result (concentration) and the raw data (absorbance values) screen.

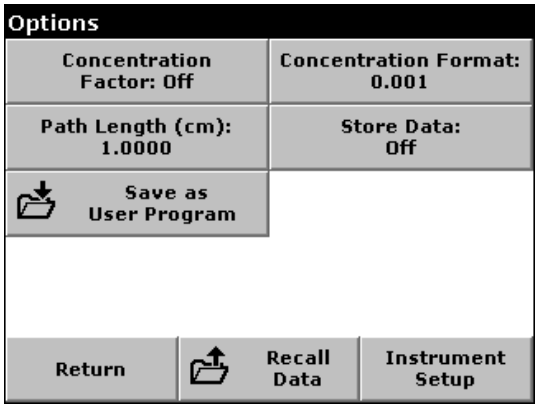
NOTE The first screen shown here (with the options menu) is the result screen while the second is the raw data screen with background correction turned on.

- **Dilution X**: lets you enter a dilution correction factor. The displayed results are corrected for dilution using the following equation:

  Con_{corr} = Conc x Dilution.



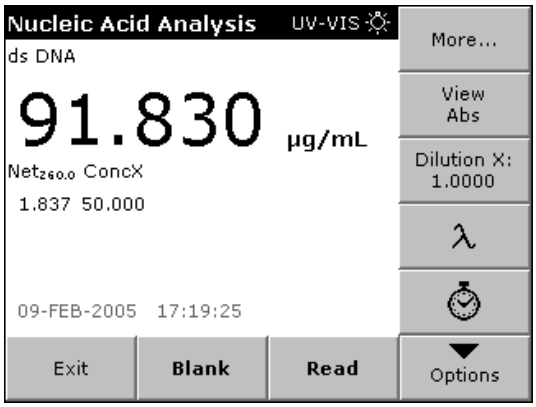
- λ: lets you modify the default wavelengths and toggle background correction on and off. When background correction is on, you can modify the default background wavelength.
- **Timer Icon**: sets the timer. Using the timer does not affect the readings that the instrument is taking. The times are not stored with sample readings.



Touch **More...** to go to the Options screen.

- **Concentration (Off/On)**: allows you to switch the concentration calculation on or off. You can also set the **Factor** and select the **Unit** in this screen.
- **Concentration Format**: sets the number of significant digits for the calculated concentration.
- **Path Length (cm)**: allows you to modify the default path length of 1 cm if you want to use small path length cells for very high sample concentrations.
- **Store Data (Off/On)**: lets you toggle the storage setting and manually store data.
- **Save as User Program**: stores the selected parameters as a user program (see "User Programs" on page 43).

dsDNA, ssDNA, and RNA

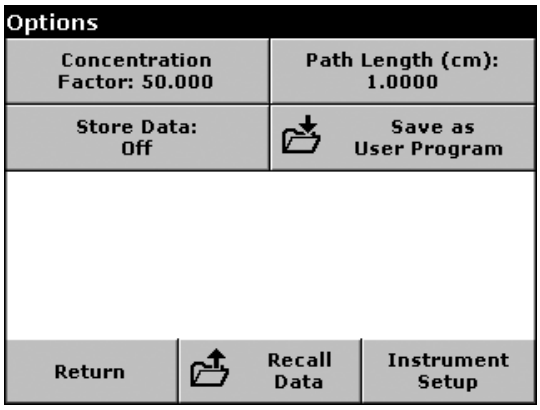


Touch **Options** for the parameter setup.

- **More...:** displays further options in the Options screen.
- **View Conc/Abs**: lets you toggle between the screen that displays the result data (concentration) and the screen that shows the raw data (absorbance values).
- **Dilution X**: lets you enter a dilution correction factor. The displayed results are corrected for dilution using the following equation:

  Con_{corr} = Conc x Dilution.

- λ: lets you modify the default wavelengths and toggle background correction on and off. When background correction is on, you can modify the default background wavelength.
- **Timer Icon**: sets the timer. Using the timer does not affect the readings that the instrument is taking. The times are not stored with sample readings.



Touch **More...** to go to the Options screen.

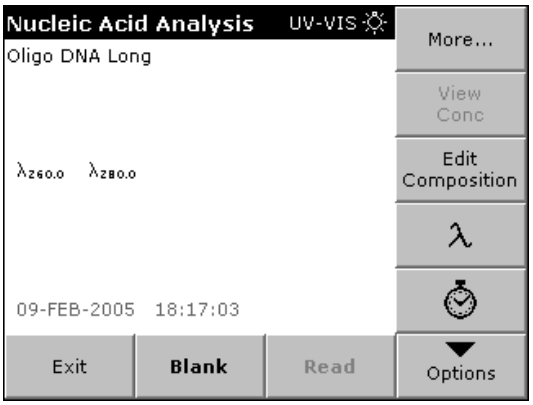
- **Concentration Factor**: allows you to modify the default factors for dsDNA, ssDNA, and RNA.

  The default factor reported the referenced literature are:

  dsDNA = 50
  ssDNA = 33
  RNA = 40

- **Concentration Format**: sets the number of significant digits for the calculated concentration.
- **Path Length (cm)**: allows you to modify the default path length of 1 cm if you want to use small path length cells for very high sample concentrations.
- **Store Data (Off/On)**: lets you toggle the storage setting and manually store data.
- **Save as User Program**: stores the selected parameters as a user program (see "User Programs" on page 43).

Oligo DNA Long and Oligo RNA Long

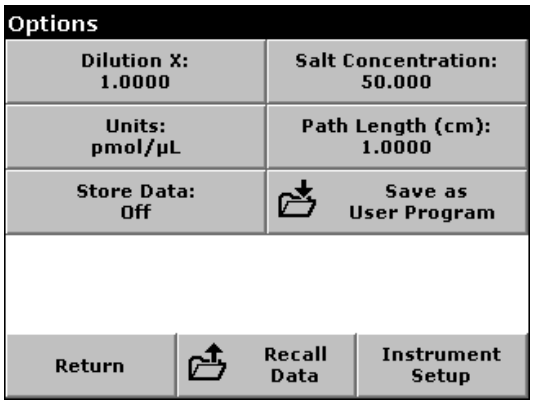


Touch **Options** for parameter setup.

- **More...**: displays further options in the Options screen.
- **View Conc/Abs**: lets you toggle between the screen that displays the results data (concentration) and the screen that shows the raw data (absorbance values).

NOTE **View Conc/Abs** option becomes enabled after you enter the base composition.

- **Edit Composition**: lets you enter the base composition (number of **A**, **C**, **G**, and **T**/**U** bases). **N** stands for unknown bases.
- λ: lets you modify the λ_2 wavelengths and toggle background correction on and off. When background correction is on, you can modify the default background wavelength.
- **Timer Icon**: sets the timer. Using the timer does not affect the readings that the instrument is taking. The times are not stored with sample readings.

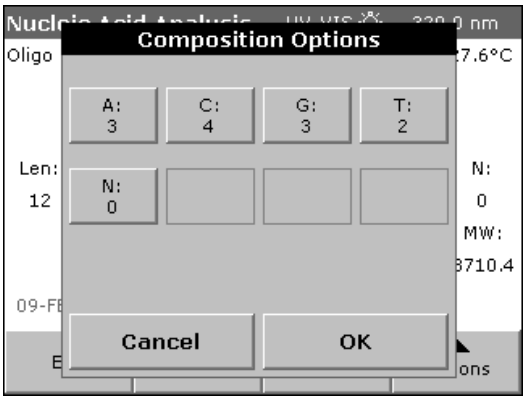


Touch **More...** to go to the Options screen.

- **Dilution X**: lets you enter a dilution correction factor. The displayed results are corrected for dilution using the following equation:

  Con_{corr} = Conc x Dilution

- **Salt Concentration**: lets you modify the default salt concentration using the numeric keypad.
- **Units**: toggle between the calculation and display of the concentration units pgmol/µL and µg/mL.
- **Path Length (cm)**: allows you to modify the default path length of 1 cm if you want to use small path length cells for very high sample concentrations.
- **Store Data (Off/On)**: lets you toggle the storage setting and manually store data.
- **Save as User Program**: stores the selected parameters as a user program (see "User Programs" on page 43).

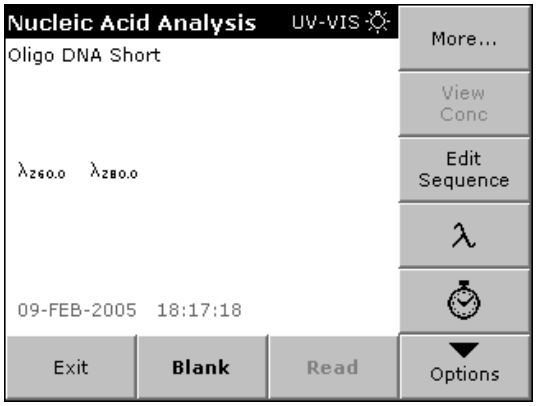


In the Composition Options screen, touch the desired base button and enter the number of bases using the numeric keypad.

NOTE For "Oligo RNA Long", the **T** (Thymine) button is replaced by the **U** (Uracil) button.

Touch **OK** when you are done.

Oligo DNA Short and Oligo RNA Short

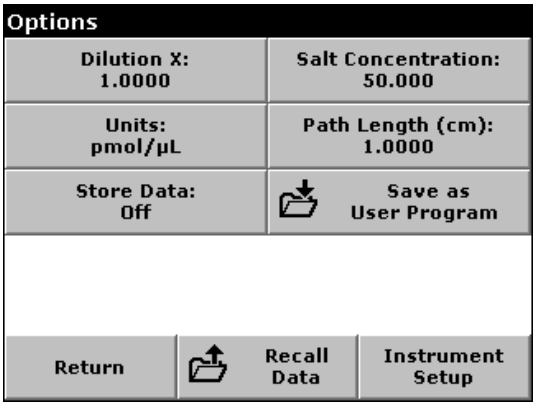


Touch **Options** for parameter setup.

- **More...:** displays further options in the Options screen.
- **View Conc/Abs**: lets you toggle between the screen that display the results data (concentration) and the screen that shows the raw data (absorbance values).

NOTE **View Conc/Abs** option becomes enabled after you enter the sequence.

- **Edit Sequence**: lets you enter the base sequence (up to 40 bases). **N** stands for unknown bases.
- λ: lets you modify the λ_2 wavelengths and toggle background correction on and off. When background correction is on, you can modify the default background wavelength.
- **Timer Icon**: sets the timer. Using the timer does not affect the readings that the instrument is taking. The times are not stored with sample readings.

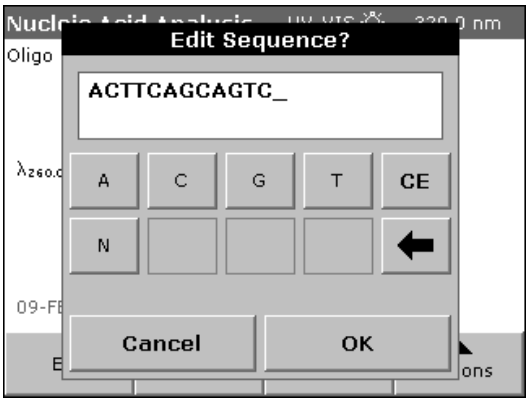


Touch **More...** to go to the Options screen.

- **Dilution X**: lets you enter a dilution correction factor. The displayed results are corrected for dilution using the following equation:

  Con_{corr} = Conc x Dilution.

- **Salt Concentration:** lets you modify the default salt concentration using the numeric keypad.
- **Units**: toggles between the calculation and display of the concentration units pgmol/µL and µg/mL.
- **Path Length (cm)**: allows you to modify the default path length of 1 cm if you want to use small path length cells for very high sample concentrations.
- **Store Data (Off/On)**: lets you toggle the storage setting and manually store data.
- **Save as User Program**: stores the selected parameters as a user program ("User Programs" on page 43).



In the Edit Sequence screen, touch the desired base buttons and enter the sequence of your sample.

- **CE**: clears the current sequence.
- **Left Arrow**: functions as a backspace button.

NOTE For "Oligo RNA Long", the **T** (Thymine) button is replaced by the **U** (Uracil) button.

Touch **OK** when you are done.

10.3 Analysis of Samples

After setting the analysis parameters, you are ready to read samples.

NOTE **Read** is disabled until a blank reading has been taken.

NOTE This description assumes that **Single Reading** is selected (default) as the reading mode and that a cell holder that holds one cell is used (the Standard Cell Holder that comes with the instrument).

For instruction on how to operate a Carousel Cell Holder, the Sipper Module, or the Peltier Temperature Control Module, see "Carousel, Sipper, and Peltier Operation" on page 117.

Before taking measurements, you need to blank the instrument using air (no cuvette in the light path) or a cuvette with a "blank" solution, which is recommended for best results.

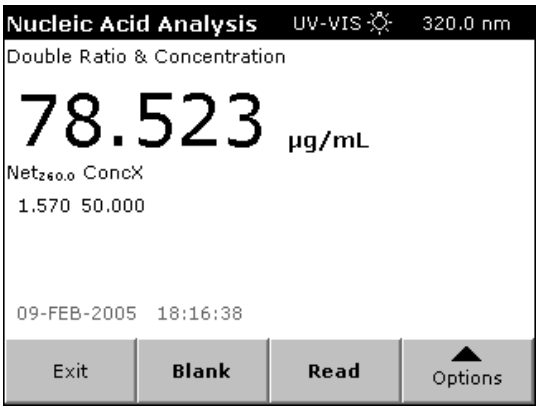
To read the samples:

1. Insert the blank cuvette into the cell holder and close the cell compartment.
2. Touch **Blank** to take the blank reading. When done, the **Read** button becomes enabled.
3. Insert a sample cuvette into the cell holder and close the cell compartment.
4. Touch **Read** to take the sample reading.

NOTE If the lid of the cell compartment is open, the instrument disables the **Blank/Read** buttons.

When trying to read a Blank or a Sample, the appropriate source must be turned on. The system assures that this is the case. You may observe the following:

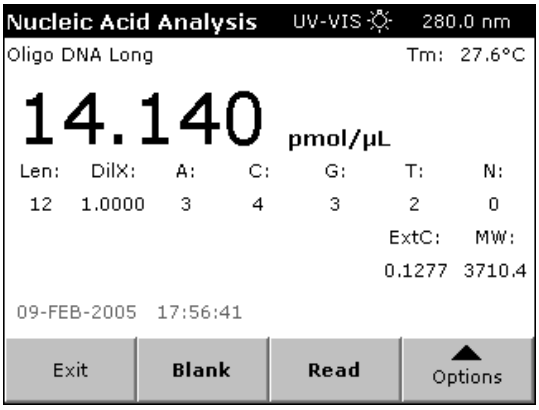
- The UV lamp automatically turns on after you have entered a wavelength in the UV spectrum. In this case, you have to wait for the UV lamp to warm up before you can take readings.
- During the warm-up phase of the UV lamp, the screen displays a "Warming up..." message and flashes the UV lamp symbol. When the UV lamp is turned on automatically, the instrument performs the reading as soon as the UV lamp is ready.



The following methods have a result screen similar to the one shown here:

- 260/280 Ratio
- Single Ratio
- Double Ratio
- dsDNA
- ssDNA
- RNA

For some of the methods, you can toggle the view to display the raw data.

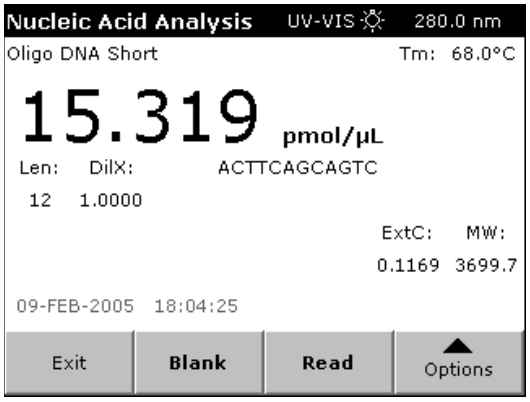


The following methods have a result screen similar to the one shown here:

- Oligo DNA Long
- Oligo RNA Long

This result screen shows the following:

- The Theoretical Melting Point (Tm), located in the top-right corner (DNA only).
- The Concentration in the selected unit, displayed in large bold characters.
- The Length (Len) of the oligo, the Dilution Factor (DilX) and the entered base composition, displayed right below the concentration.
- The Extinction Coefficient (ExtC) and the Molecular Weight (MW), displayed in the bottom-right corner of the screen.



The following methods have a result screen similar to the one shown here:

- Oligo DNA Short
- Oligo RNA Short

This result screen shows the following:

- The Theoretical Melting Point (T_m), located in the top-right corner (DNA only).
- The Concentration in the selected unit, displayed in large bold characters.
- The Length (Len) of the oligo, the Dilution Factor (DilX) and the entered sequence, displayed right below the concentration.
- The Extinction Coefficient (ExtC) and the Molecular Weight (MW), displayed in the bottom-right corner of the screen.

NOTE For instructions on how to use a Carousel Cell Holder, a Sipper, or a Peltier Temperature Controller, see "Carousel, Sipper, and Peltier Operation" on page 117.

NOTE For instructions on how to print and store data, see "Store, Recall, Send, and Delete Data" on page 125.

10.4 Calculations

The following sections describe the calculations used by methods in the Nucleic Acid Analysis mode. Each calculation provides an equation, followed by a reference number within parentheses. Where applicable, these reference numbers identify the equations described within other calculation descriptions.

NOTE The application uses the commonly used term "extinction coefficient" in place of the IUPAC term "absorptivity". These terms can be used interchangeably.

Background Correction and Net Absorbance

It is possible to correct for raised baselines, which may be caused by turbidity in the sample, using background correction. If you enable background correction, the instrument subtracts the reading at the input wavelength from the reading at the analytical wavelength(s) to calculate a net absorbance reading.

$$\text{Net Abs} = \text{Absorbance}_{\lambda} - \text{Bkg Abs} \quad (1)$$

where

$\text{Absorbance}_{\lambda}$ = Reading at the analytical wavelength
Bkg Abs = Reading at the background wavelength

Only the calculated net absorbance may be displayed instead of all the raw absorbances. If the background correction is disabled, the net absorbance is equal to the reading at the analytical wavelength.

Concentration Calculations

The 260/280 Ratio and the Single Ratio methods use the following equation to calculate the concentration:

$$\text{Conc} = \text{Net Abs} \times \text{Conc. Factor} \times \text{Dil. Factor} \quad (2)$$

The Double Ratio & Concentration, dsDNA, ssDNA, and RNA methods use the following equation to calculate the concentration:

$$\text{Conc} = \text{Net Abs} \times \text{Conc. Factor} \times \text{Dil. Factor} / \text{Pathlength} \quad (3)$$

where:

Net Abs = Net absorbance at 260 nm, calculated using equation (1)
Conc. Factor[6] = Concentration factor (user input)

Dil. Factor = Dilution factor (user input)
Pathlength = in cm (user input)

NOTE These concentrations are calculated using the net absorbance at 260 nm. However, you may change the wavelength; if you do, you must also change the concentration factor since the extinction coefficient is a function of wavelength.

Molecular Weight (Oligo DNA Long/Short)

The system calculates the molecular weight of DNA oligos from the input base composition of sequence using the following equation:

$$MW = (MW_A \times d_A) + (MW_C \times dC) + (MW_G \times dG) + (MW_T \times dT) + (MWN \times d_N) + 18.02 \quad (4)$$

where:

MW_A = Molecular weight of adenine, 313.2 g/mol
dA = Number of adenine bases (user input)
MW_C = Molecular weight of cytosine, 289.2 g/mol
dC = Number of cytosine bases (user input)
MW_G = Molecular weight of guanine, 329.2 g/mol
dG = Number of guanine bases (user input)
MW_T = Molecular weight of thymine, 304.2 g/mol
dT = Number of thymine bases (user input)
MW_N = Average molecular weight of A, C, G, T at 308.95 g/mol
dN = Number of unspecified bases (user input)

Molecular Weight (Oligo RNA Long/Short)

The system calculates the molecular weight of RNA oligos from the input base composition of sequence using the following equation:

$$MW = (MW_A \times dA) + (MW_C \times dC) + (MW_G \times dG) + (MW_U \times d_U) + (MW_N \times dN) + 18.02 \quad (5)$$

where:

MW_A = Molecular weight of adenine, 329.2 g/mol
dA = Number of adenine bases (user input)
MW_C = Molecular weight of cytosine, 305.2 g/mol
dC = Number of cytosine bases (user input)
MW_G = Molecular weight of guanine, 345.2 g/mol
dG = Number of guanine bases (user input)
MW_U = Molecular weight of uracil, 306.2 g/mol

dU = Number of uracil bases (user input)
MW_N = Average molecular weight of A, C, G, U at 321.45 g/mol
dN = Number of unspecified bases (user input)

Molar Extinction Coefficient (Oligo DNA Long)

For oligonucleotides longer than 30 bases, if the number of each type of base is known, the molar extinction coefficient (ε_{oligo}) at 260 nm, in units of L/µmol·cm, is calculated using the equation:

$$\varepsilon_{oligo} = [(\varepsilon_A \times dA) + (\varepsilon_C \times dC) + (\varepsilon_G \times dG) + (\varepsilon_T \times dT) + (\varepsilon_N \times dN)] \times 10^{-3} \quad (6)$$

where:

ε_A = Millimolar extinction coefficient of adenine, 15.4[1]
dA = Number of adenine bases (user input)
ε_C = Millimolar extinction coefficient of cytosine 7.4[1]
dC = Number of cytosine bases (user input)
ε_G = Millimolar extinction coefficient of guanine, 11.5[1]
dG = Number of guanine bases (user input)
e_T = Millimolar extinction coefficient of thymine, 8.7[1]
dT = Number of thymine bases (user input)
e_N = Average millimolar extinction coefficient of A, C, G, T at 10.75[2]
dN = Number of unspecified bases (user input)

Molar Extinction Coefficient (Oligo DNA Short)

For oligonucleotides shorter than 30 bases, the base order affects the molar extinction coefficient to the extent that the simpler formula for long oligos does not provide an accurate determination.

To calculate the molar extinction for shorter oligos, you must enter the sequence. The system calculates the molar extinction coefficient ($\varepsilon_{ABC\cdots XYX}$) at 260 nm, in units of L/µmol·cm, using the equation:

$$\varepsilon_{ABC\cdots XYZ} = [2 \times (\varepsilon_{AB} + \varepsilon_{BC} + \ldots + \varepsilon_{XY} + \varepsilon_{YZ}) - (\varepsilon_B + \varepsilon_C + \ldots + \varepsilon_X + \varepsilon_Y)] \times 10^{-3} \quad (7)$$

where:

ε_{AB} = Millimolar extinction coefficient for each base pair[1,3]

ε_{A} = Millimolar extinction coefficient for each base[1]

Molar Extinction Coefficient (Oligo RNA Long)

For oligonucleotides longer than 30 bases, if the number of each type of base is known, the molar extinction coefficient (ε_{oligo}) at 260 nm, in units of L/µmol cm, is calculated using the equation:

$$\varepsilon_{oligo} = [(\varepsilon_A \times dA) + (\varepsilon_C \times dC) + (\varepsilon_G \times dG) + (\varepsilon_U \times dU) + (\varepsilon_N \times dN)] \times 10^{-3} \quad (8)$$

where:

ε_{A} = Millimolar extinction coefficient of adenine, 15.4[1]
dA = Number of adenine bases (user input)
ε_{C} = Millimolar extinction coefficient of cytosine 7.2[1]
dC = Number of cytosine bases (user input)
ε_{G} = Millimolar extinction coefficient of guanine, 11.5[1]
dG = Number of guanine bases (user input)
ε_{U} = Millimolar extinction coefficient of uracil, 9.9[1]
dU = Number of uracil bases (user input)
ε_{N} = Average millimolar extinction coefficient of A, C, G, U at 11.0[2]
dN = Number of unspecified bases (user input)

Molar Extinction Coefficient (Oligo RNA Short)

For oligonucleotides shorter than 30 bases, the base order affects the molar extinction coefficient to the extent that the simpler formula for long oligos does not provide an accurate determination.

To calculate the molar extinction for shorter oligos, you must enter the sequence. The system calculates the molar extinction coefficient ($\varepsilon_{ABC\cdots XYX}$) at 260 nm, in units of L/µmol·cm, using the equation:

$$\varepsilon_{ABC\cdots XYZ} = [2 \times (\varepsilon_{AB} + \varepsilon_{BC} + \ldots + \varepsilon_{XY} + \varepsilon_{YZ}) - (\varepsilon_B + \varepsilon_C + \ldots + e_X + e_Y)] \times 10^{-3} \quad (9)$$

where:

ε_{AB} = Millimolar extinction coefficient for each base pair[1,3]

ε_{A} = Millimolar extinction coefficient for each base[1]

Extinction Coefficient (Oligo DNA/RNA Long/Short)

The extinction coefficient (k) at 260 nm, in units of L/µmol cm, is calculated from the molar extinction coefficient, using the equation:

$$\mathbf{k_{oligo} = \varepsilon_{oligo} \times 10^3 / Mol.\ Weight} \quad \textbf{(10)}$$

where:

ε_{oligo} = Molar extinction coefficient, calculated using equation (6), (7), (8), or (9)

Mol. Weight = Molecular weight of the oligo, calculated using equation (4) or (5)

Concentration (Oligo DNA/RNA Long/Short)

The oligonucleotide concentration is calculated in both units of pM/µL and µg/mL using the equations:

$$\mathbf{Conc\ (pM/\mu L) = (Net\ Abs \times Dil.\ Factor) / (\varepsilon_{oligo} \times Pathlength)} \quad \textbf{(11)}$$

$$\mathbf{Conc\ (\mu g/mL) = (Net\ Abs \times Dil.\ Factor) / (k_{oligo} \times Pathlength)} \quad \textbf{(12)}$$

where:

Net Abs = Net absorbance at 260 nm, calculated using equation (1)

Dil. Factor = Dilution factor, input by the user

ε_{oligo} = Molar extinction coefficient, calculated using equation (6), (7), (8), or (9)

$\mathbf{k_{oligo}}$ = Extinction coefficient, calculated using equation (10)

NOTE These concentrations are calculated using molar extinction coefficients, which were obtained from absorbance measurements at 260 nm, as described in the literature. Therefore, the wavelength used for concentration calculation is fixed at 260 nm and cannot be changed for that reason.

Melting Temperature (Oligo DNA Long)

Oligonucleotides longer than 30 bases are considered "long" oligos.

$$T_m = 81.5°C + [16.6 \times \log_{10}([salt]/1000)] - (675/L_{oligo}) + 0.41 \times \%GC) \quad (13)$$

where:

[salt]	=	Millimolar salt concentration (<1M) (user input)
$\mathbf{L_{oligo}}$	=	Number of bases in the oligonucleotide
%GC	=	GC content in the oligonucleotide, expressed as a percentage

Melting Temperature (Oligo DNA Short)

Oligonucleotides shorter or equal 30 bases are considered "short" oligos.

$$T_m = ([\Delta\Sigma H° \times 1000] / [\Delta\Sigma S° + R \times \ln(C/4)]) - 273.15 + (16.6 \times \log_{10}([salt]/1000) \quad (14)$$

where:

$\Delta H°$	=	Nearest neighbor enthalpy[5]
$\Delta S°$	=	Nearest neighbor entropy[5]
R	=	Gas constant (1.987)
C	=	Concentration from equation (11), in molar units (pMol)
[salt]	=	Millimolar salt concentration (<1M) (user input)

Where the base identity is unspecified, the calculations are performed using the average values for the pairs of nucleotides. The higher the ratio of unspecified bases to the total bases, the higher the uncertainty in the T_m calculation.

---

[1] *CRC Handbook of Biochemistry and Molecular Biology, Nucleic Acids* (1977)

[2] The value used for unspecified base is the average of the millimolar extinction coefficient for the four specified bases.

[3] The value used for unspecified base pair is the average of the millimolar extinction coefficient for all specified base pairs.

[4] Rychlik, W., Spencer, W.J., and Rhoads, R.E., Nucleic Acids Research, 18:6409 - 6412 (1990)

[5] Breslauer, K.J, Frank, R., Blöcker, H. and Marky, L.A., Proc. Natl. Acad. Sci. U.S.A., 83:3746 - 3750 (1986)

[6] The concentration factors provided in the literature generally agree that a factor of 50 should be used for double stranded DNA, a factor of 33 should be used for single stranded DNA, and a factor of 40 should be used for RNA, when data are collected at 260 nm. These factors are provided as default values. However, the user has the option of inputting any desired concentration factor.

% Dye Incorporation

The % Dye Incorporation mode performs the Analysis of Labeling Reaction for Spotted Array Sample Preparation.

11.1 Method Description

Reference:

Microarray Resource Facility
Mount Sinai School of Medicine
Protocol by Te-Hua Tearina Chu, Ph.D.

The % Dye Incorporation mode has been developed for single and duo-color methods (spotted array). It is suitable for any dye that gives similar fluorescence wavelength to Cy3 (550 nm) or Cy5 (650 nm).

Typical Procedure:

1. Use a Beckman Coulter 50 µl Microcell to analyze the entire undiluted sample.
2. Wash the microcell with water and dry it completely before use.
3. Pipette sample into microcell and place microcell into the 50 µl Microcell Holder.
4. Measure sample(s), using the % Dye Incorporation mode.
5. Pipette sample from the microcell back into the original tube.

Equations:

cDNA= (abs_{260} x volume x 37 ng/µL x 1000 pg/ng) / 324.5 pg/pmol

Cy3 = (abs_{550} x volume) / 0.15

Cy5 = (abs_{650} x volume) / 0.25

$Ratio_{Cy3}$= cDNA / Cy3

$Ratio_{Cy5}$ =cDNA / Cy5

where:

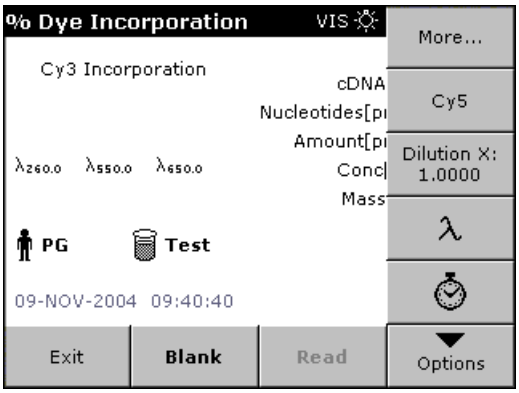
Ratio= Nucleotides/Dye Ratio
cDNA= Nucleotides (cDNA) in pmol
Cy3 = Cy3 Dye in pmol
Cy5 = Cy5 Dye in pmol
abs_{260}= Absorbance at 260 nm
abs_{550}= Absorbance at 550 nm
abs_{650}= Absorbance at 650 nm
volume= Volume in µL

1 Abs unit at 260 nm = 37 ng/µL for cDNA
324.6 pg/pmol is the average molecular weight of dNTP

NOTE >100 pmol of dye incorporation per sample and a ratio of 25-50 nucleotides / dye molecule is optimal for hybridizations.

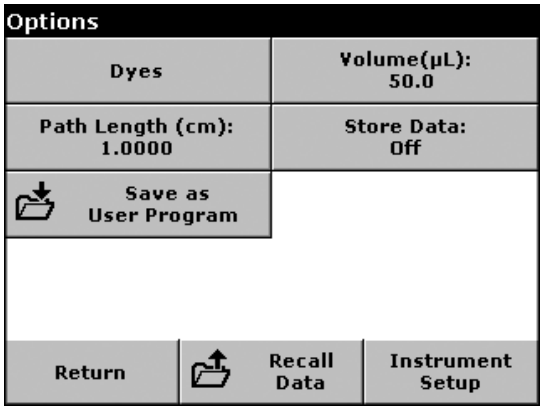
NOTE Low dye incorporation is often due to mishandled or aged Cy dyes. You need to obtain fresh dyes and repeat the sample preparation.

11.2 Parameter Setup



To enter the % Dye Incorporation mode:

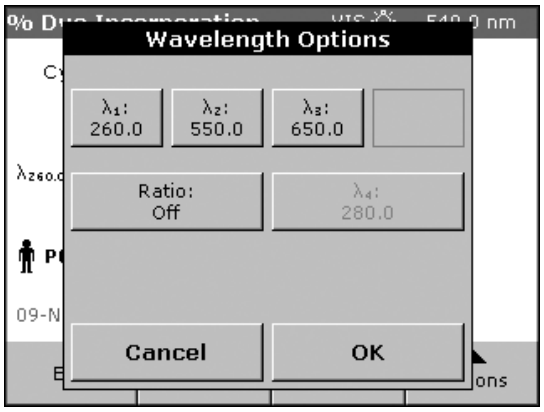
1. Touch **% Dye Incorporation** in the Main Menu.
2. Touch **Options** to set the parameters.
   - **More...**: displays further options in the Options screen.
   - **Cy3/Cy5**: toggles between the results of **Cy3** and **Cy5**.
   - **Dilution X**: lets you enter a dilution factor. The displayed concentrations are corrected for dilution using the following equation:
     $Conc_{corr}$ = Conc x Dilution
   - λ: lets you enter the wavelength(s) and allows you to turn the ratio **on** or **off**.
   - **Timer Icon**: lets you set the timer. Using the timer does not affect the readings that the instrument is taking. The times are not stored with sample readings.



3. Touch **More…** to see the previous page in the Options menu.
   - **Dyes:** lets you choose between 1 color (Cy3 or Cy5) and 2 colors (Cy3 and Cy5).

NOTE When you change the dye selection, the system resets the corresponding wavelengths for the dye(s) to its default values.

- **Volume (µL):** lets you enter the desired volume in µL.
- **Path Length (cm):** allows you to modify the default path length of 1 cm for the cuvette. The 50 µL Microcell, which is strongly recommended, has a path length of 1 cm.
- **Store Data (Off/On):** toggles the storage setting from **Off** to **On**.
- **Save as User Program:** lets you enter the number for data storage and recall (see "User Programs" on page 43).



To modify the default wavelength and/or to toggle the 260/280 ratio **On** or **Off**, touch λ to go to the Wavelength Options screen.

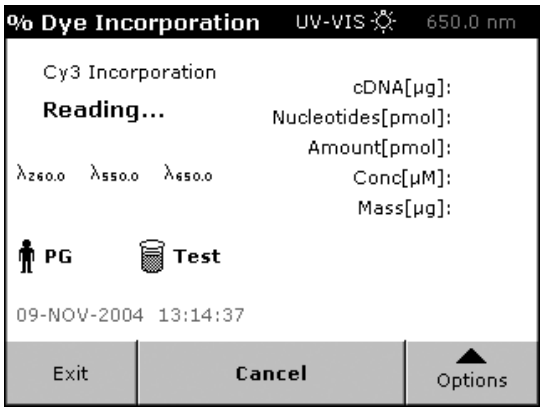
Touch λ_1 to λ_3 to change the default wavelengths settings and enter the desired wavelength using the numeric keypad.

NOTE If you chose to take readings for only one dye, the wavelength button for the other dye color is grayed out. With the selection of 2 colors, λ_2 represents the wavelength for Cy3 and λ_3 represents the wavelength for Cy5.

The **Ratio** button toggles the reporting of the 260/280 ratio between **Off** and **On**. With the ratio turned on, you can also modify λ_4, if necessary.

11.3 Analysis of Samples

After you have entered the analysis parameters, the samples are ready to be read.



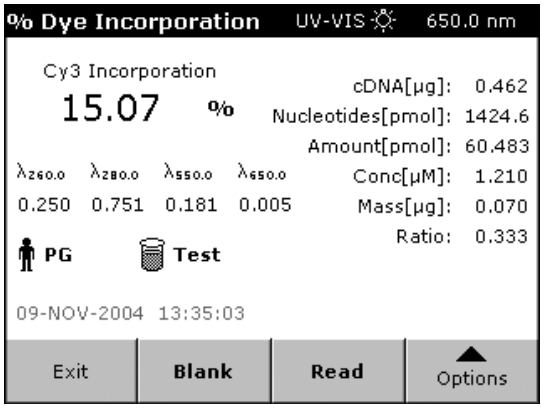
To enter the % Dye Incorporation mode:

1. Touch **% Dye Incorporation** in the Main Menu.
2. Insert a cuvette with a blank solution into the cell holder and close the cell compartment (to blank against air, leave the "Blank" cell position empty).

NOTE If the lid of the cell compartment is open, the instrument disables the **Blank/Read** buttons.

3. Touch **Blank** to blank the instrument. The screen displays the "Blanking..." message in the bottom-left corner of the screen.
4. When the system has been blanked, insert a sample cuvette into the cell holder and close the cell compartment.
5. Touch **Read** to start the reading. The screen displays the "Reading..." message in the bottom-left corner of the screen and then reports the results for the sample.

NOTE During the reading process, the screen replaces the **Read/Blank** buttons with a **Cancel** button. You may touch **Cancel** to stop the reading and discard the data.

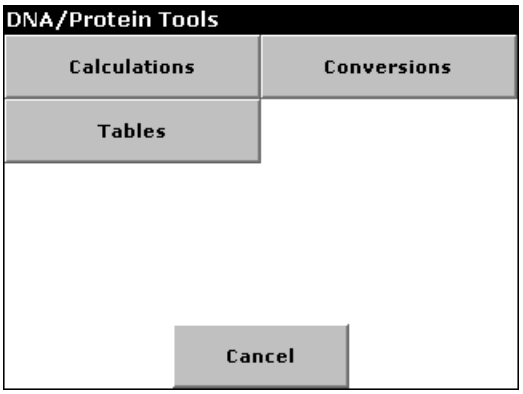


On this results screen, the 260/280 Ratio has been turned **On** and the ratio is reported together with % Dye Incorporation results.

This screen shows the results for one color only. To see the results for the second color (Cy5), touch **Options** and then **Cy5**. Selecting this option toggles the result screens between **Cy3** and **Cy5**.

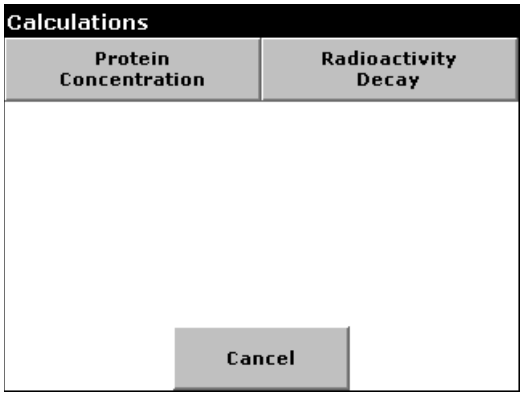
DNA/Protein Tools Mode

The DNA/Protein Tools do not measure samples. Instead, these tools or functions are available to help you with your daily laboratory tasks. The tools provide calculations, common conversions, and a number of common tables from the microbiology handbook.



To enter the DNA/Protein Tools mode, touch **DNA/Protein Tools** in the Main Menu.

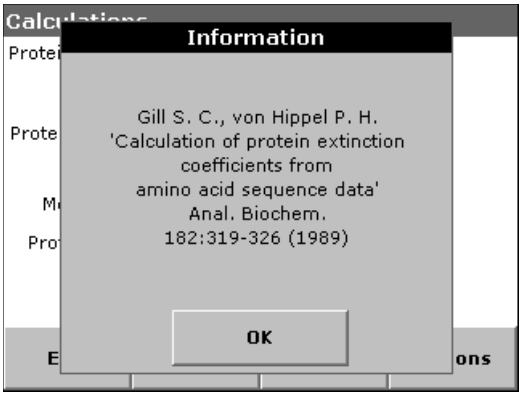
12.1 Calculations



In the Calculations screen, touch the desired button to perform the function.

Touch **Cancel** to return to the DNA/Protein Tools screen.

NOTE When entering a function, a results screen without values may appear. When this happens, touch **Options** first and enter or change the parameters before touching **Calculate** to calculate and display the result(s).



Most functions provide additional information, such as references or calculation descriptions.

When extra information is available, the **Info** button is enabled. To access this information, touch **Info**.

Protein Concentration from Amino Acid Sequence Data

Determining specific absorptivity constants[1] (a_s) and the concentration for synthetic or recombinant proteins or peptides is a necessary component of biopharmaceutical production and formulation. Once an a_s value is determined for a particular protein in a well-defined formulation matrix, the concentration of that protein in solution can be rapidly determined from its measured absorbance and the specific absorptivity (or extinction) constant using a derivative of Beer's law.

This function of the DNA/Protein Tools calculates the following parameters:

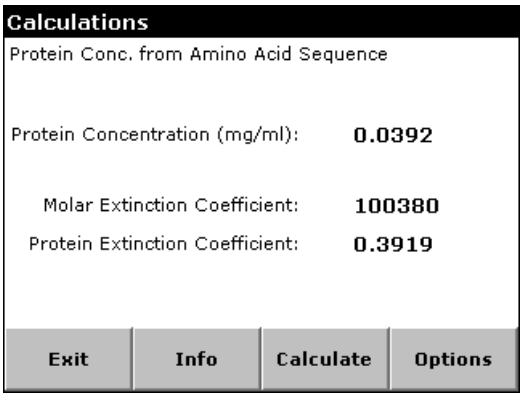
- Molar Extinction Coefficient (or Absorptivity Constant)
- Protein Extinction Coefficient
- Protein Concentration in mg/ml

The calculation is based on the following input parameters:

- # of Thyrosines
- # of Tryptophans
- # of Cysteines (or Cystines)
- Molecular Weight of the Protein
- Absorbance at 280 nm
- Dilution Factor

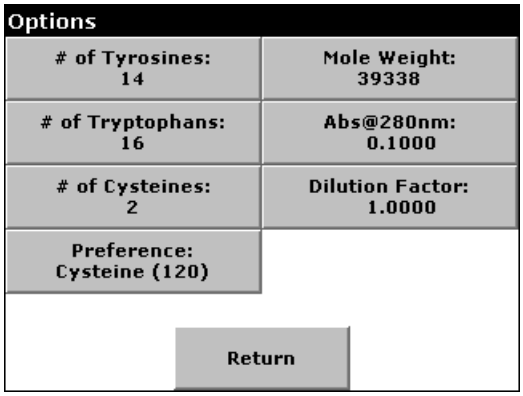
NOTE The absorbance measurement must be taken separately.

[1]Gill S.C., von Hippel P.H. "Calculation of protein extinction coefficients from amino acid sequence data". Anal. Biochem. 182:319-326(1989)



To enter the mode:

1. Touch **Protein Concentration** in the Calculations menu.
2. Touch **Info** to display the available information for this function.
3. Touch **Calculate** to perform the calculation and display the result(s) based on the parameters set in Options.



4. Touch **Options** to set the parameters for the calculation.
5. Touch the parameter that identifies the value you want to change and set a parameter either by:
   - using a numeric keypad to enter a value or
   - selecting the radio button that displays the desired value.

NOTE The **Preference** button allows you to choose the factor for Cysteine (which is 60) or Cystine (which is 120).

Radioactive Decay Correction

When working with radiolabeled DNA probes or other radiolabeled assays, you may need to correct for the radioactive decay.

This function of the DNA/Protein Tools performs a half-life correction for the radioactive substance used in the assay or experiment.

The following parameters must be supplied:

- Original Date
- Original Activity (in µCurie, DPM, or kBq)
- Current Date
- Half-life of the Isotope*

* For ^{125}I, ^{131}I, ^{32}P, ^{33}P, ^{35}S, and ^{3}H, the isotope is selected and the system automatically supplies the respective half-life time. For other isotopes, please enter the corresponding half-life time for the isotope.

Equation:

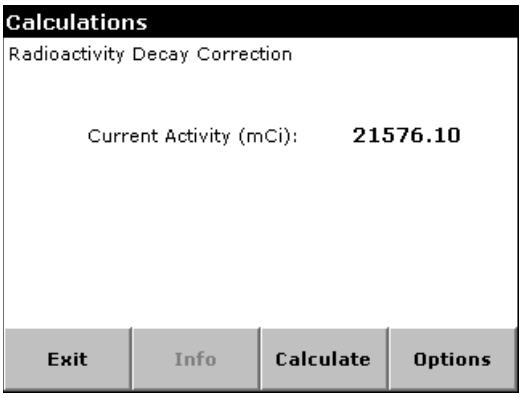
$$act_{corr} = act_{curr} - (act_{curr} \times (1 - Exp(-0.693 \times act_{days} / h_{time})))$$

where:

act_{corr} = Corrected Activity
act_{curr} = Current Activity
act_{days} = Difference in days
h_{time} = Half-life in days

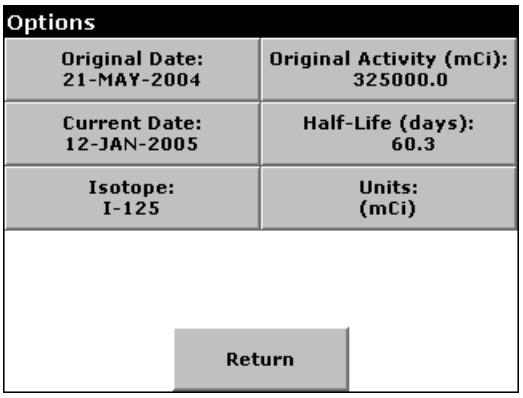
Half-lives of Stored Isotopes:

^{125}I = 60.30 Days
^{131}I = 8.10 Days
^{32}P = 14.28 Days
^{33}P = 25.40 Days
^{35}S = 187.40 Days
^{3}H = 4490 Days or 12.3 Years

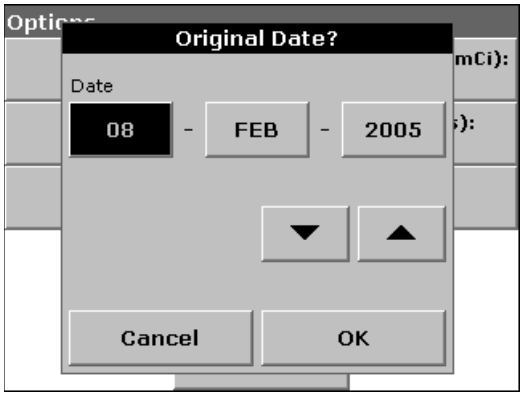


To enter this function:

1. Touch **Radioactivity Decay** in the Calculations menu.
2. Touch **Calculate** to perform the calculation and display the result(s) based on the parameters set in Options.



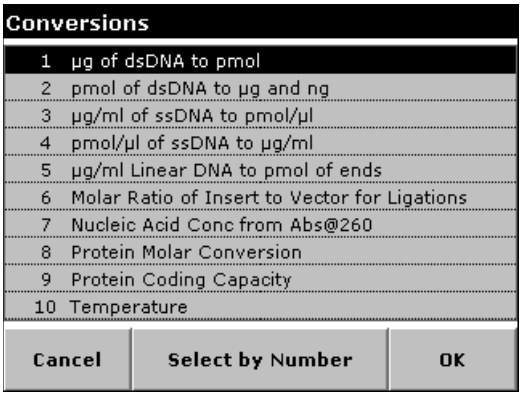
3. Touch **Options** to set the parameters for the calculation.
4. Touch the parameter that identifies the value you want to change and set a parameter either by:
   - using a numeric keypad to enter a value or
   - selecting the radio button that displays the desired value.



To select a date:

1. Touch the button you want to change (day, month, or year).
2. Use the up and down arrows to select the desired value.
3. Touch **OK**.

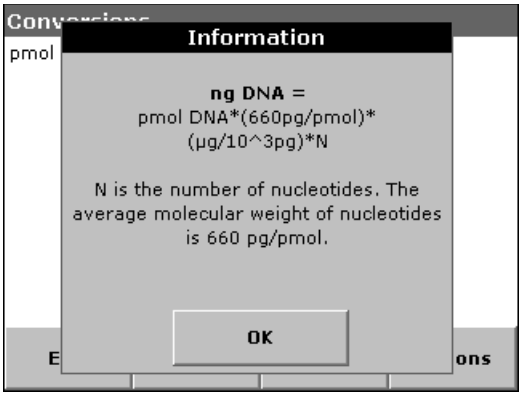
12.2 Conversions


Conversions
1 µg of dsDNA to pmol
2 pmol of dsDNA to µg and ng
3 µg/ml of ssDNA to pmol/µl
4 pmol/µl of ssDNA to µg/ml
5 µg/ml Linear DNA to pmol of ends
6 Molar Ratio of Insert to Vector for Ligations
7 Nucleic Acid Conc from Abs@260
8 Protein Molar Conversion
9 Protein Coding Capacity
10 Temperature
Cancel
Select by Number
OK

In the Conversions screen:

1. Touch the desired line to highlight it or touch **Select by Number** and enter the desired number.
2. Touch **OK** to perform the function.
3. Touch **Cancel** to return to the DNA/Protein Tools screen.

NOTE When entering a function, the instrument may display a result screen without values. When this happens, touch **Options** first and enter or change the parameters before you touch **Calculate** to calculate and display the result(s).



Most functions provide additional information, such as references or calculation descriptions.

When extra information is available, the **Info** button is enabled. To access this information, touch **Info**.

Micrograms or dsDNA to Picomoles

This function converts the amount of dsDNA (µg) to pmol.

The following parameters must be supplied:

- Length of DNA (in base pairs)
- Amount of dsDNA (in µg)

Equation:

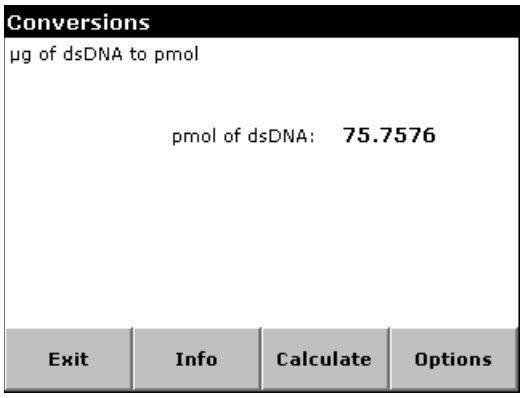
$$\mathbf{DNA_{pmol} = DNA_{\mu g} \times (pmol/660pg) * (10^6 pg/\mu g) / (1/N)}$$

where:

$\mathbf{DNA_{pmol}}$ = Result (dsDNA in pmol)
$\mathbf{DNA_{\mu g}}$ = dsDNA in µg
N = Number of nucleotides

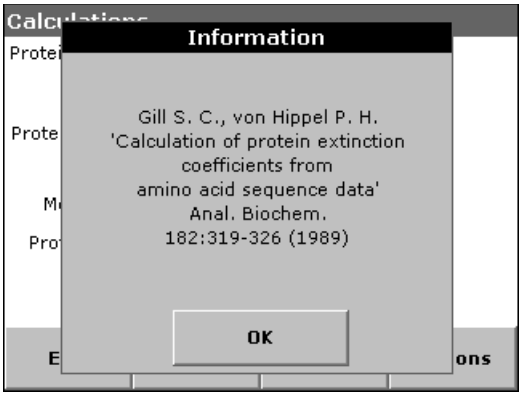
The average molecular weight of nucleotides is 660 pg/pmol.

Example for Conversion Functions



To enter a function:

1. Select **µg of dsDNA to pmol** or the desired function in the Conversions menu.
2. Touch **Info** to display the available information for this function.
3. Touch **Calculate** to perform the conversion and display the result(s) based on the parameters set in Options.



4. Touch **Options** to set the parameters for the calculation.
5. Touch the parameter that identifies the value you want to change and set a parameter either by:
   - using a numeric keypad to enter a value or
   - selecting the radio button that displays the desired value.

NOTE Operation of the other functions in this section is similar.

Picomoles of dsDNA to Micrograms and Nanograms

This function converts the amount of dsDNA (pmol) to µg and ng.

The following parameters must be supplied:

- Length of DNA (in base pairs)
- Amount of dsDNA (in pmol)

Equation:

$$\mathbf{DNA_{\mu g} = (DNA_{pmol} \times 660 pg/pmol \times N) / (\mu g/10^6 pg) \times N}$$

$$\mathbf{DNA_{ng} = (DNA_{pmol} \times 660 pg/pmol \times N) / (\mu g/10^3 pg) \times N}$$

where:

$\mathbf{DNA_{\mu g}}$	=	Result (dsDNA in $_{\mu g}$)
$\mathbf{DNA_{ng}}$	=	Result (dsDNA in $_{ng}$)
$\mathbf{DNA_{pmol}}$	=	dsDNA in pmol
N	=	Number of nucleotides

The average molecular weight of nucleotides is 660 pg/pmol.

Micrograms/mL of Oligonucleotides (ssDNA) to Picomoles/µL

This function converts oligonucleotide (ssDNA) concentrations (µg/ml) to pmol/µL.

The following parameters must be supplied:

- Length of Oligo (in nucleotides)
- Concentration (in µg/ml)

Equation:
$Oligo_{pmol/µl} = (Oligo_{µg/ml}$ x (ml/1000µl) / (pmol/330pg) x (10^6pg/µg) x (1/N)

where:

$Oligo_{pmol/µl}$ = Result (Oligo in pmol/µl)
$Oligo_{µg/ml}$ = Oligo in µg/ml
N = Number of nucleotides

The average molecular weight of nucleotides is 330 pg/pmol.

Picomoles/µL of Oligonucleotides (ssDNA) to Micrograms/mL

This function converts Oligonucleotide (ssDNA) concentrations (pmol/µL) to µg/ml.

The following parameters must be supplied:

- Length of Oligo (in nucleotides)
- Concentration (in pmol/µl)

Equation:
$Oligo_{µg/ml} = (Oligo_{pmol/µl}$ x (1000µl/ml) / (330pg/pmol) x (µg/10^6pg) x N

where:

$Oligo_{µg/ml}$ = Result (Oligo in µg/ml)
$Oligo_{pmol/µl}$ = Oligo in pmol/µl
N = Number of nucleotides

The average molecular weight of nucleotides is 330 pg/pmol.

Micrograms of Linear DNA to Picomoles of Ends

This function converts DNA sizes (kb) to DNA amounts.

The following parameters must be supplied:

- Amount of DNA (in µg)
- Size of DNA (in kb)

Equation:

$$\mathbf{Ends_{pmol} = DNA_{\mu g} \times (pmol/660pg) \times (10^6 pg/\mu g) \times (1/N) \times 2 \times (kb/1000bp)}$$

$\mathbf{Ends_{pmol}}$ = Result (Ends in pmol)
$\mathbf{DNA_{\mu g}}$ = Linear DNA in µg
$\mathbf{DNA_{kb}}$ = Number of nucleotides in kilobases (kb)

The average molecular weight of a single nucleotide pair is 660 pg/pmol.

The number of ends in a linear DNA molecule is 2 and the conversion factor from kilobases to base pairs is kb/1000 bp.

Molar Ratio of Insets to Vector for Ligations

This function calculates the ng of insert for a 1:1 molar ratio.

The following parameters must be supplied:

- Length of Insert (in kb)
- Length of Vector (in kb)
- Amount of Vector (in ng)

Equation:

$$\mathbf{insert_{ng} = insert_{kb} / vector_{kb} \times vector_{amt}}$$

where:

$\mathbf{insert_{ng}}$ = Result (Insert in ng)
$\mathbf{insert_{kb}}$ = Insert Length in kilobases (kb)
$\mathbf{vector_{kb}}$ = Vector Length in kilobases (kb)
$\mathbf{vector_{amt}}$ = Vector Amount in kilobases (kb)

Generally ligations are tested at 1:3, 1:1, and 3:1 insert:vector molar ratios.

Nucleic Acid Concentration from Abs@260

This function estimates the DNA/RNA/Oligo concentration from an absorbance value.

NOTE You may also use the Nucleic Acid Analysis mode of the DU 730 to measure the sample and obtain the result (estimated concentration) at the same time.

The following parameters must be supplied:

- Absorbance at 260 nm
- Type of DNA (dsDNA, ssDNA, RNA, or Oligo)

Equation:
conc = abs_{260} x factor

where:

conc = Result (Concentration)
abs_{260} = Absorbance at 260 nm
factor = Concentration Factor (see listing below)

Common concentration factors:

dsDNA = 50
ssDNA = 35
RNA = 40
Oligo = 20

Protein Molar Conversion

This function performs a couple of molar conversion for proteins.

The following parameters must be supplied:

Parameter Set #1:

- Protein Size (in kDa)
- Amount of Protein (in pmol)
  = Converts to µg of Protein

or Parameter Set #2:

- Protein Size (in kDa)
- Amount of Protein (in µg)
  = Converts to pmol of Protein

or Parameter Set #3:

- Amount of Protein (in pmol)
- Amount of Protein (in µg)
  = Converts to kDa (Protein Size)

Equations:

Parameter Set #1:
$$\mathbf{prot_{\mu g} = (size_{kDa} \times prot_{pmol}) \times 10^{9} \times (1 / 10^{12})}$$

Parameter Set #2:
$$\mathbf{prot_{pmol} = (prot_{\mu g} / size_{kDa}) \times (1 / 10^{9}) * 10^{12}}$$

Parameter Set #3:
$$\mathbf{size_{kDa} = (prot_{\mu g} / prot_{pmol}) \times (1 / 10^{9}) * 10^{12}}$$

where:

$\mathbf{size_{kDa}}$	=	Protein Size in kDa
$\mathbf{prot_{pmol}}$	=	Protein Amount in pmol
$\mathbf{prot_{\mu g}}$	=	Protein Amount in µg

Protein Coding Capacity

This function encodes Amino Acids, calculates the size of the DNA, and predicts the size and length of the protein.

One of the following parameters must be supplied:

- DNA Size (in bases)
  = Converts to Protein Size
  = Converts to Protein Length
- or Protein Size (in kDa)
  = Converts to DNA Size
  = Converts to Protein Length
- Protein Length (in Amino Acids)
  = Converts to DNA Size
  = Converts to Protein Size

Equation:

$\mathbf{dna_{prot}}$ **= input x 3**
$\mathbf{prot_{dna}}$ **= input x / 3**
$\mathbf{prot_{dna}}$ **= input x 0.11**

where:

$\mathbf{dna_{size}}$ = Size of DNA (bases)
$\mathbf{prot_{size}}$ = Size of Protein (kDa)
$\mathbf{prot_{length}}$ = Length of Protein (amino acids)

To encode Amino Acids, divide the size of the DNA (in bases) by 3.

To calculate the size of the DNA, multiply the number of amino acids by 3.

The predicted size of the protein is the number of amino acids times 0.11 kDa. 0.11 kDa is the average molecular weight of an amino acid.

Temperature Conversion

This function provides temperature conversions.

The following parameters must be supplied:

- Temperature in Fahrenheit, Celsius, or Kelvin
  = Converts to Fahrenheit, Celsius, or Kelvin

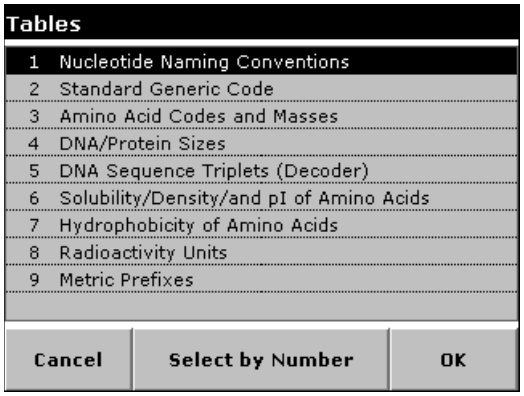
Equation:

$t_C = 0.55556 \times (t_F - 32)$
$t_F = (1.8 \times t_C) + 32$
$t_K = (0.55556 \times (t_F - 32)) + 273.16$
$t_F = (1.8 \times (t_K - 273.16)) + 32$
$t_K = t_C + 273.16$
$t_C = t_K - 273.16$

where:

t_F = Temperature in Fahrenheit
t_C = Temperature in Celsius
t_K = Temperature in Kelvin

12.3 Tables



In the Tables screen:

1. Touch the desired line to highlight it or touch **Select by Number** and enter the desired number.
2. Touch **OK** to perform the function.
3. Touch **Cancel** to return to the DNA/Protein Tools screen.

NOTE Tables do not perform a function. They simply provide information.

Nucleotide Naming Conventions

This function provides a table with a number of nucleotide translation conventions:

IUB/GCG = International Union of Biochemistry convention translates sequences for GENBANK, University of Wisconsin Genetic Computing Group, EMBL, and the National Biomedical Research Foundation.

Staden = The Staden convention is used by the Cambridge University database.

Sanger = The Sanger convention is currently not used.

Stanford = The Stanford convention is used by the Stanford University database.

Standard Genetic Code

This function provides the table of the standard genetic code.

Amino Acid Codes and Masses

This function provides a table with the names of the 20 common amino acids, the single-letter code, the corresponding three-letter abbreviation as well as its mass.

DNA/Protein Sizes

This function provides a table with conversion information for DNA and Proteins.

DNA Sequence Triplets (Decoder)

This function provides a table with the common amino acid names and its DNA Sequence Triplets (Decoder) also called the DNA Codons as they are read on the sense (5' to 3') strand of DNA.

Solubility, Density, and pI of Amino Acids

This function provides a table with the common amino acid names and list the solubility (g/100g at 25ºC), the Crystal Density (g/ml), and the isoelectric point (pI) at 25ºC.

Hydrophobicity of Amino Acids

This function provides a table that indicates hydrophobicity of amino acids, based on the following references: Janin (1979), Wolfenden, Kyte & Doolittle, and Rose.

Radioactivity Units

This function provides a table with radioactivity units and the corresponding conversion information.

Metric Prefixes

This function provides a table with metric prefixes.

Carousel, Sipper, and Peltier Operation

13.1 Carousel Holder

For information on changing sample modules, loading and inserting the carousel, see "Carousel Holder (Sample Changer)" on page 18. For details on activating the Carousel Holder and setting the carousel options, see "Carousel and Module Options" on page 40.

The table below lists the activated/disabled conditions of **Blank**, **Read**, and **Blank & Read** buttons under various Carousel settings.

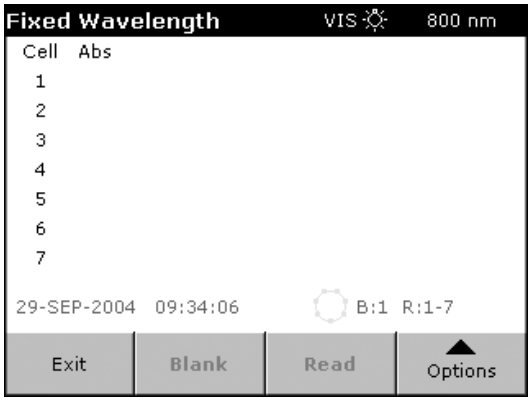
Table 13.1 Shifting Activated/Disabled Conditions for Carousel Settings

Carousel Mode	Initial State (Display)	After Blanking (Display)
B: 1 R: 1 – n	[Blank] [Read]	[Read] [Read]
B: 1 – n R: 1 – n	[Blank] [Read]	[Blank] [Read]
B: 1 R: 2 – n	[Blank & Read]	[Blank & Read]
B: 1, (3), (5) R: 2, (4), (6)	[Blank & Read]	[Blank & Read]

B = "Blank" cell position
R = "Sample" cell position
***n* = number of samples in the Carousel.**

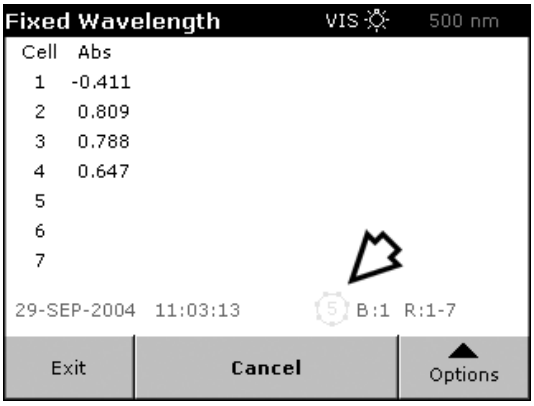
Fixed Wavelengths Measurements

This section applies to Fixed Wavelength, SCA, Protein Assay Analysis, Nucleic Acid Analysis, and % Dye Incorporation modes.



Touch **Blank** (or **Blank** & **Read**) to start the first set of measurements.

The status bar displays the instrument operation: "Reading... (Blanking)", while the Carousel turns and the instrument takes readings as programmed through the Instrument Setup menu.



After starting the "Reading....." sequence, the screen replaces the **Blank/Read** buttons with a **Cancel** button.

The instrument lists each reading on the display as it is taken. When blanking or reading, the position number appears in the Carousel icon (see arrow).

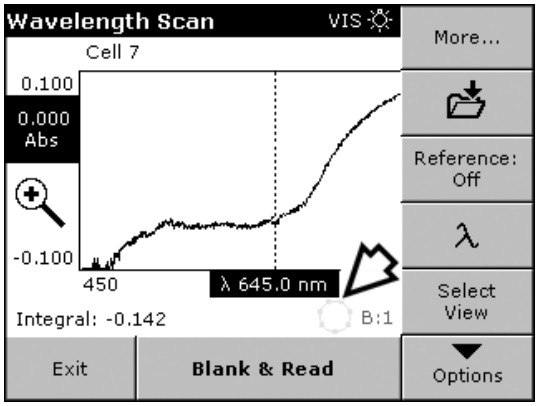
NOTE Touch **Cancel** at any time to start over and to erase all readings from the current set of measurements.

After finishing the reading sequence the Carousel turns back to the initial position. The screen reactivates the **Blank/Read** buttons.

Wavelength Scan

NOTE You may call the Carousel Options screen at any time to set the Carousel options in an instant by touching the Carousel Icon (see arrow). Alternatively, you can go through the **Options** button for this function.

NOTE Entering the mode or changing scanning parameters, requires a new baseline scan (or blank scan).



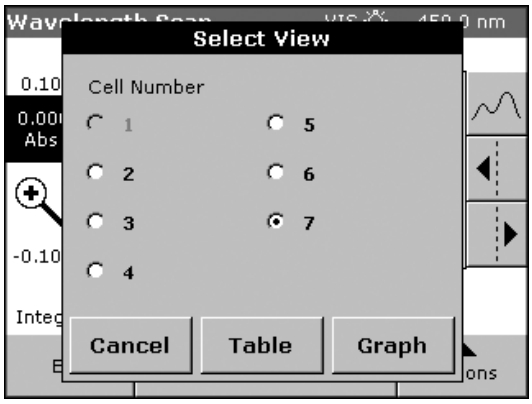
After performing the baseline scan, the instrument is ready to scan samples.

After starting the "Reading..." sequence, the screen replaces the **Blank/Read** buttons with a **Cancel** button.

NOTE Touch **Cancel** at any time to start over and erase all scans from the current set of measurements.

You may touch the Carousel Icon (see arrow) to change the Carousel Settings in an instant (without returning to the Instrument Setup menu).

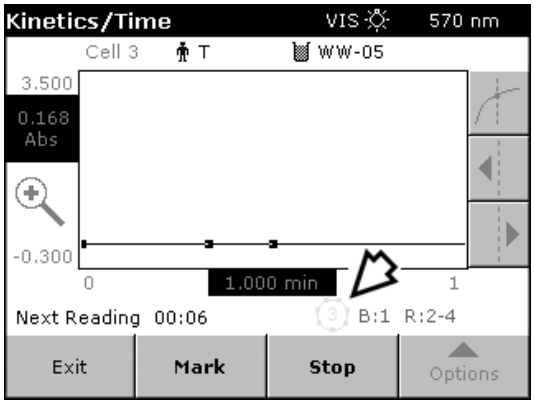
After finishing the reading sequence the Carousel turns back to the initial position. The screen reactivates the **Blank/Read** buttons.



To display a graph (scan) or table (tabular data) from the available scans, touch **Options** and then **Select View**.

Select the **Cell Number** by touching a radio button and then touch **Table** or **Graph** to display the scan data in tabular form or its graphical representation.

Kinetics/Time



After blanking the system, the instrument is ready to start the kinetics run.

After starting the "Reading..." sequence, the screen replaces the **Blank/Read** buttons with a **Mark** and a **Stop** button.

The **Mark** button enables you to place a marker (a small black square) at the current point as the instrument displays a line while reading the sample.

NOTE Touch **Stop** at any time to start over and erase all readings from the current set of measurements.

You may touch the Carousel Icon (see arrow) to change the Carousel Settings in an instant (without returning to the Instrument Setup menu).

After finishing the reading sequence (run), the Carousel turns back to the initial position. The screen reactivates the **Blank/Read** buttons. This completes the kinetic run.

Readings in all defined carousel positions are taken at each time interval. For example, the 6 defined carousel positions provide 6 kinetic runs.

Touch **Options** and **Select View** to select the kinetic run you want to display and analyze.

13.2 Sipper Module

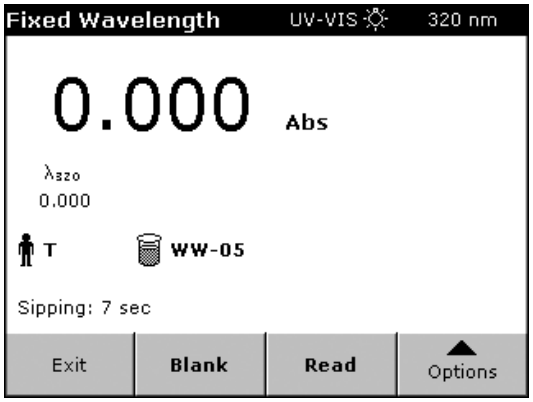
For information on changing sample modules, loading and inserting the Sipper Module, or setting the sipper parameters, see "Sipper Module" on page 19.

In general, the operation is similar to measurements with a "one cuvette" cell holder. The only difference is that, with the sipper, you don't have to change cuvettes. The blank and sample solutions are sipped into the cell (a "flow cell") and then returned or pumped to waste.

NOTE Make sure to route the drain tube to an appropriate drain or collection vessel when disposing the sample.

Fixed Wavelengths Measurements

This section applies to Fixed Wavelength, SCA, Protein Assay Analysis, Nucleic Acid Analysis, and % Dye Incorporation modes.



Place the sample inlet tube into the blank solution and touch **Blank** or **Read** button.

Leave the inlet tube in the blank solution or the sample vessel until the Sipper pump stops and the settling cycle begins.

After starting the sipping/reading sequence, the screen replaces the **Blank/Read** buttons with a **Cancel** button. At any time, you can erase all readings from the current set of measurements and start over by touching **Cancel**.

The screen displays the remaining time in seconds in the bottom-right corner of the screen, in place of the date.

NOTE If you need to stop the sip cycle, touch **Cancel**. During the sipping and reading process, the screen replaces the **Blank/Read** buttons with a **Cancel** button. The instrument skips the sip and settle cycles.

When the settling cycle is complete, the screen displays the **Blank** button.

- If the purge is set up for a manual start the instrument waits for you to touch **Purge**.
- In the automatic mode, the purge begins immediately after the reading without operator intervention.

At this time, if the sample is going to waste, you can place a demineralized water purge at the sample inlet to rinse the sample cell between readings.

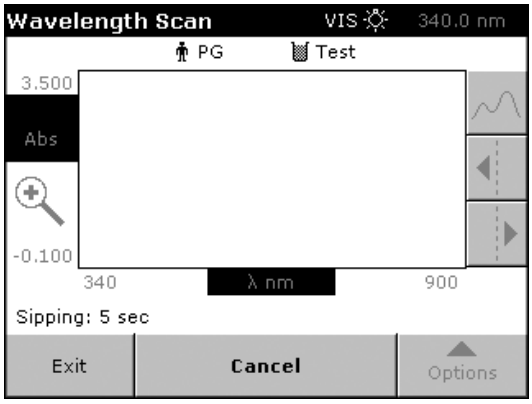
Leave the inlet tube in the purge solution until the Sipper pump stops.

The screen displays the remaining time in seconds in the bottom-right corner of the screen, in place of the date.

NOTE If you need to stop the purge cycle during the purge interval, touch **Cancel**.

When the sample purge is complete, the instrument is ready for the next sample.

Wavelength Scan



Place the sample inlet tube into the blank solution and touch **Blank** or **Read**.

Leave the inlet tube in the blank solution or the sample vessel until the Sipper pump stops and the settling cycle begins.

After starting the sipping/reading sequence, the screen replaces the **Blank/Read** buttons with a **Cancel** button.

NOTE If you need to stop the sip cycle, touch **Cancel**. The instrument skips the sip and settle cycles.

The screen displays the remaining time in seconds in the bottom-right corner of the screen, in place of the date.

When the settling cycle is complete, the screen displays the **Blank** button.

- If the purge is set up for a manual start the instrument waits for you to touch **Purge**.
- In the automatic mode, the purge begins immediately after the reading without operator intervention.

At this time, if the sample is going to waste, you can place a demineralized water purge at the sample inlet to rinse the sample cell between readings.

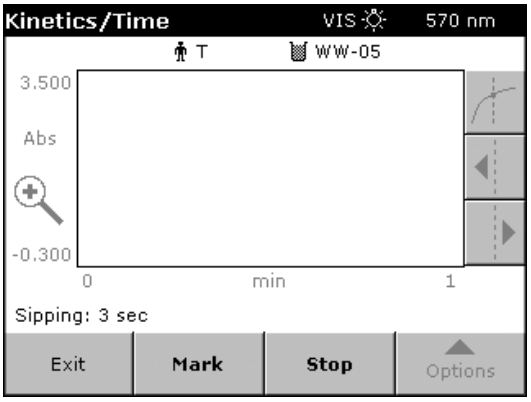
Leave the inlet tube in the purge solution until the Sipper pump stops.

The screen displays the remaining time in seconds in the bottom-right corner of the screen, in place of the date.

NOTE If you need to stop the purge cycle during the purge interval, touch **Cancel**.

When the sample purge is complete, the instrument is ready for the next sample.

Kinetics/Time



Place the sample inlet tube into the blank solution and touch **Blank** or **Read**.

Leave the inlet tube in the blank solution or the sample vessel until the Sipper pump stops and the settling cycle begins.

After starting the sipping/reading sequence, the screen replaces the **Blank/Read** buttons with a **Mark** and a **Stop** button.

NOTE If you need to stop the sip cycle, touch **Stop**. The instrument skips the sip and settle cycles.

The screen displays the remaining time in seconds in the bottom-right corner of the screen, in place of the date.

When the settling cycle is complete, the screen displays the **Blank** button.

- If the purge is set up for a manual start, the instrument waits for you to touch **Purge**.
- In the automatic mode, the purge begins immediately after the reading without operator intervention.

At this time, if the sample is going to waste, you can place a demineralized water purge at the sample inlet to rinse the sample cell between readings.

Leave the inlet tube in the purge solution until the Sipper pump stops.

The screen displays the remaining time in seconds in the bottom-right corner of the screen, in place of the date.

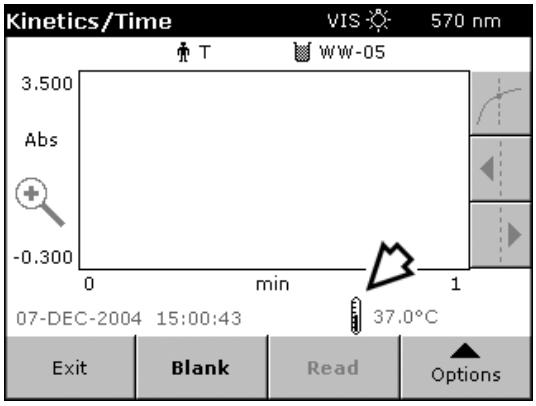
NOTE If you need to stop the purge cycle during the purge interval, touch **Cancel**.

When the sample purge is complete, the instrument is ready for the next sample.

13.3 Peltier Module

For information on changing sample modules, loading and inserting the Peltier Temperature Control Module, or setting the temperature parameters, see "Peltier Temperature Control Module" on page 25.

All Modes



With the Peltier Temperature Control module installed, the instrument behaves just like a system with a regular Single Cell Holder.

However, with the module installed, a Thermometer Icon and the current temperature appears at the bottom-right corner of the screen.

You may touch the Thermometer Icon (see arrow) to change the Peltier Settings in an instant (without returning to the Instrument Setup menu).

Store, Recall, Send, and Delete Data

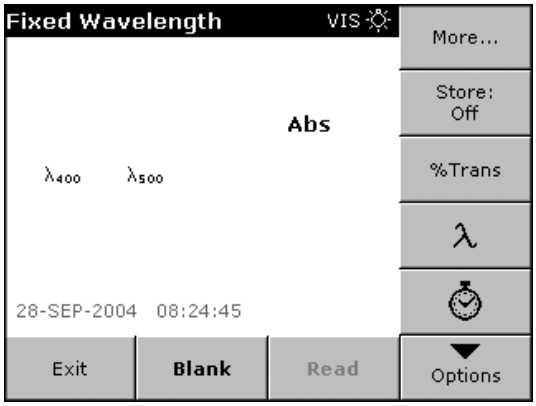
14.1 Storing Data

DU 700 Series instruments can store up to 1000 readings taken in the most modes (Fixed Wavelength, SCA, Protein Assay Analysis, Nucleic Acid Analysis, and % Dye Incorporation). In Wavelength Scan mode, the instrument can store up to 20 scans. The Kinetics/Time mode stores up to 20 kinetic runs.

Storing Data - Auto/Manual

This section applies to Fixed Wavelength, SCA, Protein Assay Analysis, Nucleic Acid Analysis, and % Dye Incorporation modes.

You can set the instrument to store the data automatically (**Store: On**), or store data manually at your discretion (**Store: Off**).



Single Reading Mode

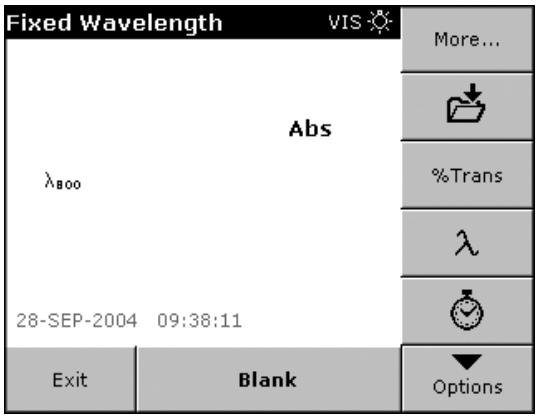
When the Options menu displays the **Store: Off** button, the instrument does not automatically store readings.

To print the data or save it to a memory device, the readings must be stored until the set of data is complete. The data can then be printed or sent to a memory device using the Data Recall function. Therefore, it is recommended to store your data.

To store specific data manually, set the **Store:** button to **Off.** After taking the reading, set the **Store:** button to **On** to store the reading/result. The system briefly displays the message: "Data stored". You must then toggle back to **Store: Off,** otherwise, the consequent readings are stored automatically.

To store data automatically, toggle the button to **Store: On** (automatic storage). When you then touch **Read**, the instrument reads the sample, places the reading/result automatically in the data log, and briefly displays the message: "Data stored".

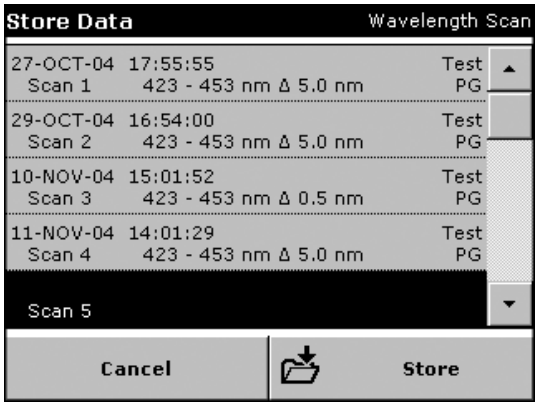
NOTE When the instrument's memory (data log) is full, the instrument automatically deletes the oldest data, allowing space to store new data.



Continuous Reading Mode

You can touch the Store icon (folder with arrow) in the Options menu at any time to place the current reading/result into the data log. The system briefly displays the message: "Data stored".

Storing Scans (Wavelength Scan)


Store Data
Wavelength Scan
27-OCT-04 17:55:55 Test
Scan 1 423 - 453 nm Δ 5.0 nm PG
29-OCT-04 16:54:00 Test
Scan 2 423 - 453 nm Δ 5.0 nm PG
10-NOV-04 15:01:52 Test
Scan 3 423 - 453 nm Δ 0.5 nm PG
11-NOV-04 14:01:29 Test
Scan 4 423 - 453 nm Δ 5.0 nm PG
Scan 5
Cancel
Store

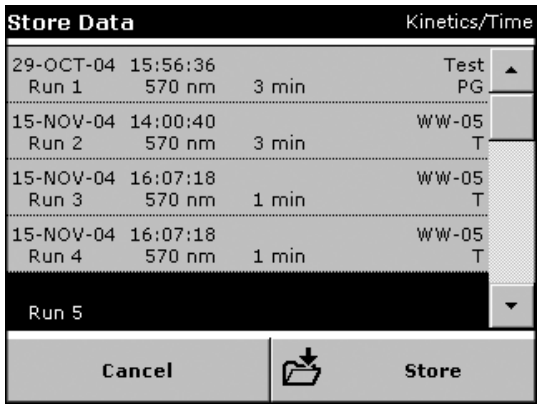
After the scan(s) have been taken, touch the **Options** button and then the Store icon (folder with arrow).

The Store Data screen displays. Touch the line (Scan #) where you want to store the first, and maybe only, scan and then touch the **Store** button (with the folder icon) to place the scan(s) in storage.

NOTE After taking multiple scans, the instrument stores the scans sequentially, starting with the highlighted scan #. If there are scans already stored in subsequent locations, the storage location will be overridden. After reaching the maximum storage location (20), the instrument stores the remaining scans, starting with scan #0. In this case, stored scans will be overridden.

The system briefly displays the message: "Data stored" and returns to the previous screen. You can touch **Cancel** to return to the previous screen without storing the scan(s).

Storing Kinetic Data (Kinetics/Time)



After completing the kinetic run, touch the **Options** button and then the Store icon (folder with arrow).

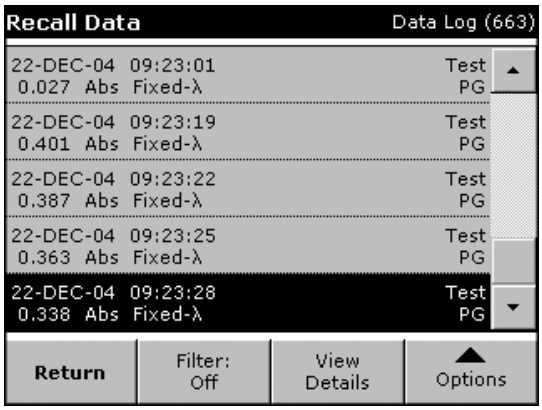
The Store Data screen displays. Touch the line (Run #) where you want to store the actual run and then touch the **Store** button (with the folder icon) to place the kinetic data in storage.

The system briefly displays the message: "Data stored" and returns to the previous screen. You can touch **Cancel** to return to the previous screen without storing the kinetic data.

14.2 Recalling Data, Scans, and Kinetic Runs

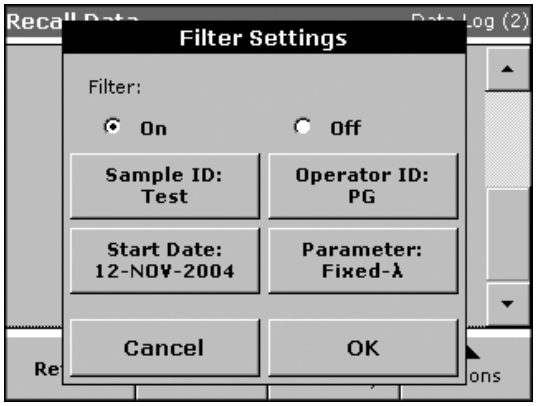
Recalling Data

This section applies to Fixed Wavelength, SCA, Protein Assay Analysis, Nucleic Acid Analysis, and % Dye Incorporation modes.



To recall stored data, touch **Recall Data** from the Main Menu and then **Data Log**. The screen displays a list of stored data, if available, as shown on the left.

When you touch **View Details**, the screen displays complete information for the selected sample. The button changes to **View Summary** to let you return to the previous screen.

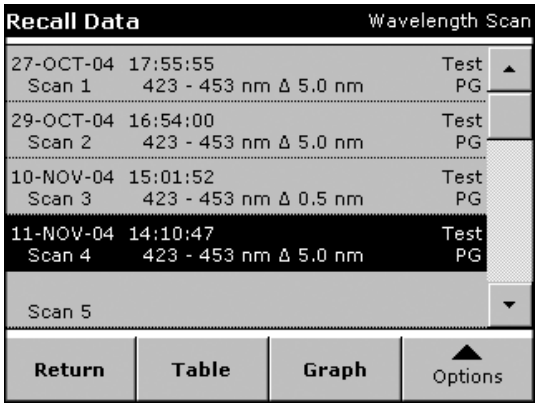


To filter-out desired data, use the **Filter: On/Off** button. With the filter activated, you may select any combination of the following options:

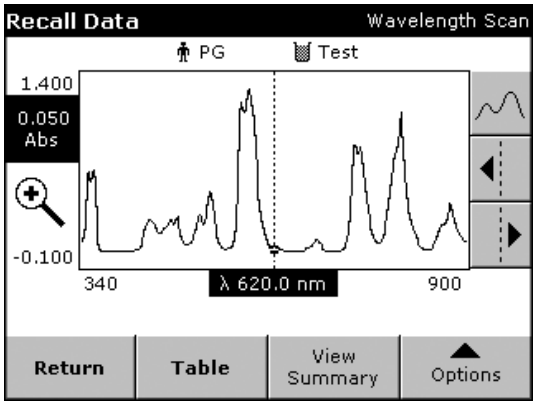
- Sample ID
- Operator ID
- Start Date
- Parameter

Touch **OK** to filter the stored data based on the settings.

Recalling Scans (Wavelength Scan)


Recall Data
Wavelength Scan
27-OCT-04 17:55:55 Test
Scan 1 423 - 453 nm Δ 5.0 nm PG
29-OCT-04 16:54:00 Test
Scan 2 423 - 453 nm Δ 5.0 nm PG
10-NOV-04 15:01:52 Test
Scan 3 423 - 453 nm Δ 0.5 nm PG
11-NOV-04 14:10:47 Test
Scan 4 423 - 453 nm Δ 5.0 nm PG
Scan 5
Return
Table
Graph
Options

To recall stored scans, touch **Recall Data** from the Main Menu and then **Wavelength Scan**. The screen displays a list of stored scans, if available, as shown on the left.

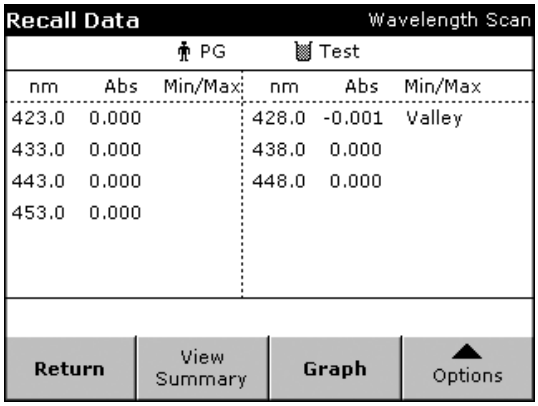


To display the graphical representation of the selected scan (example shown on the left), touch **Graph**.

The button changes to **View Summary** to let you return to the previous screen.

If manual scaling has been selected, you can use the alphanumeric keypad to set the limits y_{min} and y_{max}. In this case, the graph adjusts to display only the scan values in the selected range.

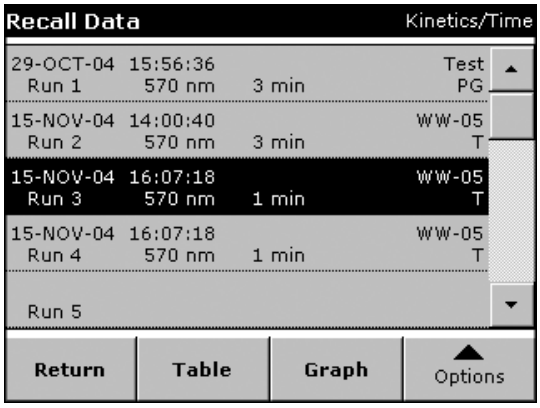
If automatic scaling has been selected, the system sets the limits automatically to display the total scan range.



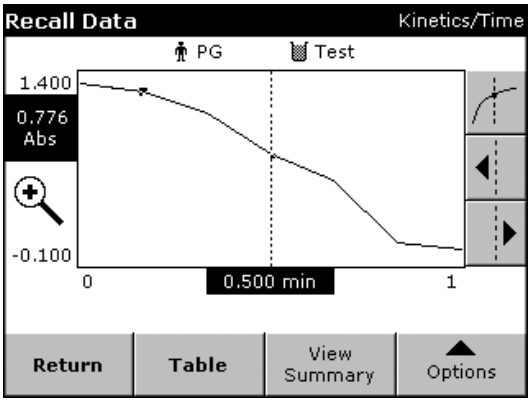
To display the tabulated data of the selected scan (example shown on the left), touch **Table**.

The button changes to **View Summary** to let you return to the previous screen.

Recalling Kinetic Data (Kinetics/Time)

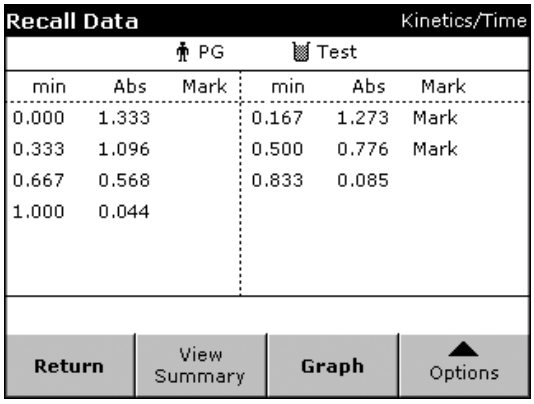


To recall kinetic data, touch **Recall Data** from the Main Menu and then **Kinetics/Time**. The screen displays a list of stored kinetic runs, if available, as shown on the left.



To display the graphical representation of the kinetic run (example shown on the left), touch **Graph**.

The button changes to **View Summary** to let you return to the previous screen.



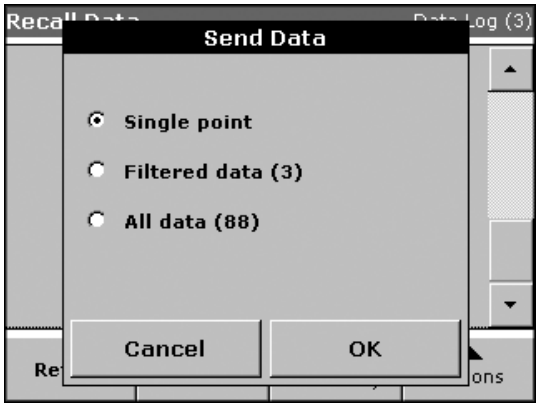
To display the tabulated data of the selected kinetic run (example shown on the left), touch **Table**.

The button changes to **View Summary** to let you return to the previous screen.

14.3 Sending Data, Scans, and Kinetic Runs

Sending Data

This section applies to Fixed Wavelength, SCA, Protein Assay Analysis, Nucleic Acid Analysis, and % Dye Incorporation modes.



You can send data to a printer or a memory device using the available USB ports. Since the system automatically senses connected devices, it sends the data to all available devices.

To send data:

1. Touch the **Options** button.
2. Touch **Send Data** (USB Port icon). The Send Data screen displays, enabling you to send a single point (selected item), the filtered data, or all data to the desired destination(s) by touching the respective radio button.
3. Touch **OK** to send the data.

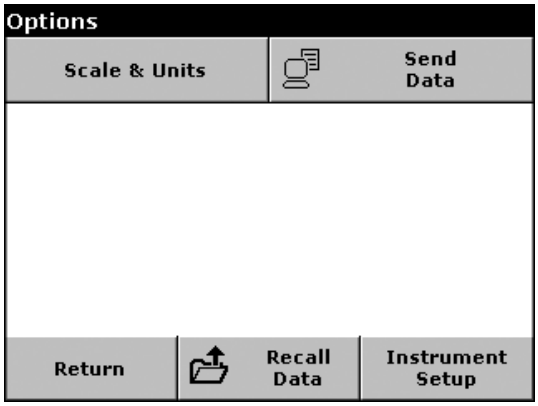
NOTE The number in brackets shows the total number of data assigned to this selection.

The instrument automatically sends the data as a single DATALOG file to the memory device using the CSV (Comma Separated Value) file format.

The file name uses the following format: DL*Year_Month_Day_Hour_Minute_Second*.csv

NOTE CSV files are the preferred format for data import into spreadsheet and graphing programs as well as statistical packages.

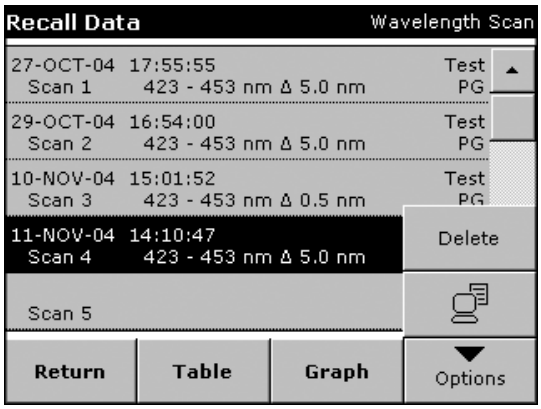
Sending Scan Data (Wavelength Scan)



You can send scan data to a printer or a memory device using the available USB ports. Since the system automatically senses connected devices, it sends the data to all available devices.

To send scan data directly from the Wavelength Scan mode:

1. Touch **Options**.
2. Touch **More...**.
3. Touch **Send Data** (USB Port icon). The instrument immediately sends to the connected devices the data of all scans that currently reside in memory.



To send scan data from the Recall Data screen:

1. Highlight the line with the desired scan #.
2. Touch **Options**.
3. Touch **Send Data** (USB Port icon). The instrument immediately sends to the connected devices the data of the selected scan.

NOTE You may repeat this procedure for the remaining scan data you want to send to the target location(s).

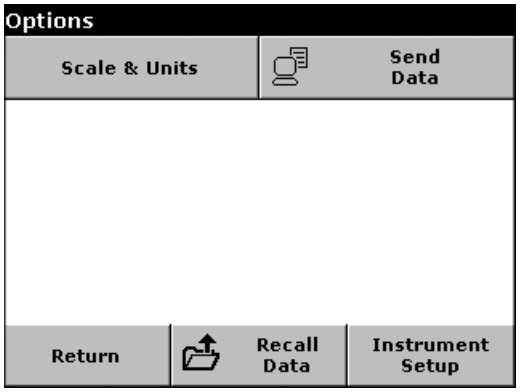
The instrument automatically sends the data as a single WLDATA file to the memory device using the CSV (Comma Separated Value) file format.

The file name uses the following format:

ScanData_*X*.csv
where
X is the Scan # (1-20)

NOTE CSV files are the preferred format for data import into spreadsheet and graphing programs as well as statistical packages.

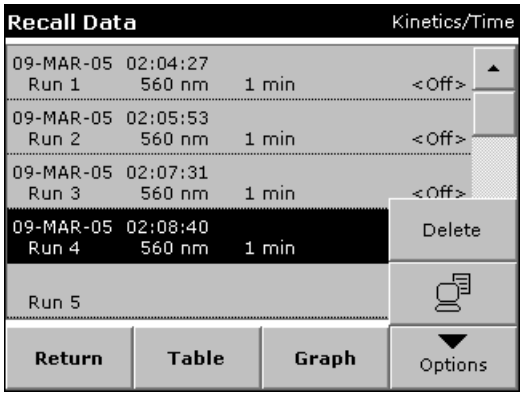
Sending Kinetic Data (Kinetics/Time)



You can send kinetic data to a printer or a memory device using the available USB ports. Since the system automatically senses connected devices, it sends the data to all available devices.

To send kinetic data directly from the Kinetics/Time mode:

1. Touch **Options**.
2. Touch **More...**.
3. Touch **Send Data** (USB Port icon). The instrument sends the actual kinetic data immediately to the connected devices.



To send kinetic data from the Recall Data screen:

1. Highlight the line with the desired Run #.
2. Touch **Options**.
3. Touch **Send Data** (USB Port icon). The instrument immediately sends the kinetic data of the selected run to the connected devices.

   The instrument automatically sends the data as a single TCDATA file to the memory device using the CSV (Comma Separated Value) file format.

   The file name uses the following format:

   TCData_*X*.csv
   where
   X is the Scan # (1-20)

NOTE CSV files are the preferred format for data import into spreadsheet and graphing programs as well as statistical packages.

14.4 Deleting Data, Scans, and Kinetic Runs

Deleting Data

This section applies to the Fixed Wavelength, SCA, Protein Assay Analysis, Nucleic Acid Analysis, and % Dye Incorporation modes.

To delete stored data:

1. Touch **Options**.
2. Touch **Delete**. The Delete Data screen displays.

3. Choose whether you want to delete a single point (selected item), the filtered data, or all data by touching the respective radio button.

4. Touch **OK** to delete the data.

NOTE The number in brackets shows the total number of data assigned to this selection.

Deleting Scan Data (Wavelength Scan)

To delete stored scan data (from the Recall Data screen):

1. Highlight the line with the Scan # you want to delete.
2. Touch **Options**.
3. Touch **Delete**. The instrument immediately deletes the scan data in the highlighted storage location.

Deleting Kinetic Data (Kinetics/Time)

To delete kinetic runs (from the Recall Data screen):

1. Highlight the line with the Run # you want to delete.
2. Touch **Options**.
3. Touch **Delete**. The instrument immediately deletes the kinetic data in the highlighted storage location.

15.1 Cleaning Requirements

Spectrophotometer

- Do not expose the instrument to extremes of temperature (such as heating, direct sunlight, or other sources of heat).
- All-round unobstructed ventilation of the instrument must be assured at all times.
- The instrument must not be operated or kept in extremely dusty, damp, or wet locations.
- Keep the surface of the instrument, the cell compartment, and all accessories clean and dry at all times. Splashes or spills on and in the instrument should be cleaned up immediately.
- Clean the enclosure, the cell compartment, and all accessories with a soft damp cloth. A mild soap solution can also be used.
- Dry the cleaned parts carefully with a soft cotton cloth.

Display

- Take care not to scratch the display. Do not touch the screen with ball pens, pencils, or similar pointed objects.
- Clean the display with a soft, lint-free, oil-free cotton cloth. Diluted window cleaner liquid can also be used.

WARNING Under no circumstances should the instrument or the display or the accessories be cleaned with solvents such as white spirit, acetone, etc.

Cell Cleaning Instructions

Cell Description

Beckman Coulter cells for UV/Visible spectrophotometers have two optical and two non-optical facing sides. The decal on one of the non-optical surfaces identifies the type of material used for the optical windows. A blue decal indicates silica windows, which transmit in the UV and visible regions from 190 to 2500 nm. A green decal indicates special optical glass windows, which transmit in the visible region only, from 320 to 2500 nm.

Always position the cell in the spectrophotometer, so that the light is transmitted through the optical windows. A higher than expected absorbance reading indicates that the cell may have been inserted in the wrong orientation.

Cell Cleaning Instructions

1. Rinse the cell with a mild solvent (distilled water for aqueous solutions, an organic solvent for organic materials) as soon as possible after each use.

WARNING Do not allow the solvent or sample solution to evaporate from the cell. Evaporation usually leaves a deposit on the cell window, which can require extensive cleaning.

2. If the cell cannot be cleaned immediately, soak the cell in distilled water. Do not soak the cell in a detergent solution, as it can etch the cell windows.

3. Clean the cell with a mild detergent. The detergent must be a true solution and must not contain particulate matter. Dilute (10%) Trace-Klean solution, available from Beckman, is recommended. Rinse the cell immediately with distilled water. Never let the detergent remain in the cell. Never use a brush or other instrument, which might scratch the sides of the cell.

CAUTION Non-dilute Trace-Klean is a highly alkaline solution and should be handled with care.

4. To remove protein deposits, soak the cell in dilute Trace-Klean solution at 58ºC for 15 minutes. Then rinse with distilled water.

5. For other hard-to-remove deposits, wash with a solution made from equal parts by volume ethanol and 3N HCL. Rinse immediately with distilled water. Do not soak the cell in this solution for more than 30 seconds.

CAUTION Do not use highly alkaline solutions, abrasive materials, or hot concentrated acids to clean the cell.

6. Dry the cell by inverting it and placing it in a cell rack. For faster drying, use a vacuum. Do not blow the cell dry with air, unless the air is known to be clean. Most laboratory air sources have minute oil particles, originating from the compressor pump.

NOTE If the cell is to be used immediately, rather than drying the cell completely, rinse the cell with the next sample solution, then fill the cell.

The outside surfaces of the cell may be dried and lightly buffed using lens tissue or a soft, lint-free cloth.

Cell Deterioration

There are two main causes of cell deterioration, both of which result in reduced transmission through the cell.

- Films - which are deposited by solvent evaporation, strong wetting agents, or inadequate cleaning. These decrease the transmission properties of the cell and may cause contamination of subsequent samples.
- Etching - caused by the continued use of strong alkali or concentrated mineral acids, either in the sample or in the cleaning solution. Etching is also caused by weak alkaline solutions left in the cell for long periods of time. Etched cells have reduced transmission properties because light is scattered from the etched surfaces.

To detect the extent of cell deterioration, measure the transmittance versus air at 220, 240, and 270 nm (silica cells), or 320nm (optical glass cells). A new Beckman cell filled with double-distilled water will have a transmittance of at least 80% (0.1 abs.).

Caution should be observed when using different cells for the blank (or reference) and the sample, especially if the cells have deteriorated. Fill all the cells with the same solution and verify that the readings are sufficiently similar before recording sample results.

For a complete description of the Beckman cell line, refer to the *Supplies and Consumables Catalog*, Bulletin 8500A. In the United States call 800-742-2345. Outside the United States contact the local Beckman office.

Flowcell Maintenance/Cleaning Instructions

Flowcell Cleaning Instructions

Flowcells exhibit unique cleaning problems due to the difficulty in cleaning the interior surfaces. Experience has shown that most problems with Flowcells are due to the cells not being clean. Dirty cells cause air bubbles to be entrapped, may cause errors due to reduced transmission, and may contaminate the sample.

It is important that samples and cleaning agents not be left in a Flowcell for long periods of time. When not in use the cell should be flushed and filled with distilled or deionized water. If this is not done, substances will be deposited on the interior surfaces, which may be very difficult to remove.

Also, a Flowcell should never be allowed to air dry. It should either be kept filled with distilled water or dried with pure bottled air or dry nitrogen.

Use only spectral grade alcohol and pure distilled or deionized water when cleaning or flushing Flowcells. Solutions may be recycled during cleaning procedures but fresh solution should always be used for finial rinses.

Conditioning a new Flowcell

A new Flowcell or a Flowcell that has not been used recently needs to be conditioned. Conditioning will produce an even, smooth flow of sample solution through the Flowcell and help prevent bubbles from forming.

To condition a Flowcell, rinse with deionized water, flush with air, and then rinse with spectral grade methanol. Flush with air again, and then rinse with deionized water. The Flowcell is now ready to use.

Cleaning Procedure - Daily

1. Flush with 1 mL of deionized water.
2. Flush with 2 mL of 10% Trace-Klean solution.

WARNING Concentrated Trace-Klean is a highly alkaline solution and should be handled with care. Always wear protective goggles to protect eyes against splashing liquid. In case of eye contact, immediately flush with running water for 15 minutes.

3. Flush with a minimum of 5 mL of deionized water.
4. Leave the Flowcell filled with deionized water.

NOTE Before next use, flush with methanol, then with deionized water.

Cleaning Procedure - Contaminated Flowcells

This procedure should be followed if bubbles remain in the Flowcell, if the Flowcell is known to be dirty, if the results are not reproducible, or if the results appear to be inaccurate.

1. Flush with deionized water for 2 minutes minimum.
2. Flush with methanol for 2 minutes minimum.
3. Flush with deionized water for 1 minute.
4. Flush with methanol for 2 minutes minimum
5. Flush with deionized water for 1 minute.
6. Flush with 10% Trace-Klean solution for 2 minutes minimum. (See NOTE below.)
7. Flush with deionized water for 1 minute.
8. Leave the Flowcell filled with deionized water.

NOTE To remove protein deposit from the inside of the Flowcell, rinse the Flowcell with a 10% Trace-Klean solution at 56ºC for 15 minutes.

Cleaning Procedure - Severely Contaminated Flowcells

Occasionally a Flowcell may become severely contaminated with substances, which have dried on the interior surfaces. Standard cleaning procedures may not clean these Flowcells. To clean them, flush with 50% hydrochloric acid (6 normal) for ten seconds then follow immediately with a 1-minute flush using deionized water. Follow this with the cleaning procedure, described in "Cleaning Procedure - Contaminated Flowcells" on page 140.

WARNING When handling hydrochloric acid, always use protective clothing and chemical safe goggles to prevent skin and eye contact. Ensure that the working area is well ventilated to avoid inhalation of vapors. In case of contact, immediately wash affected area with running water for 15 minutes. For eyes, also contact a physician immediately.

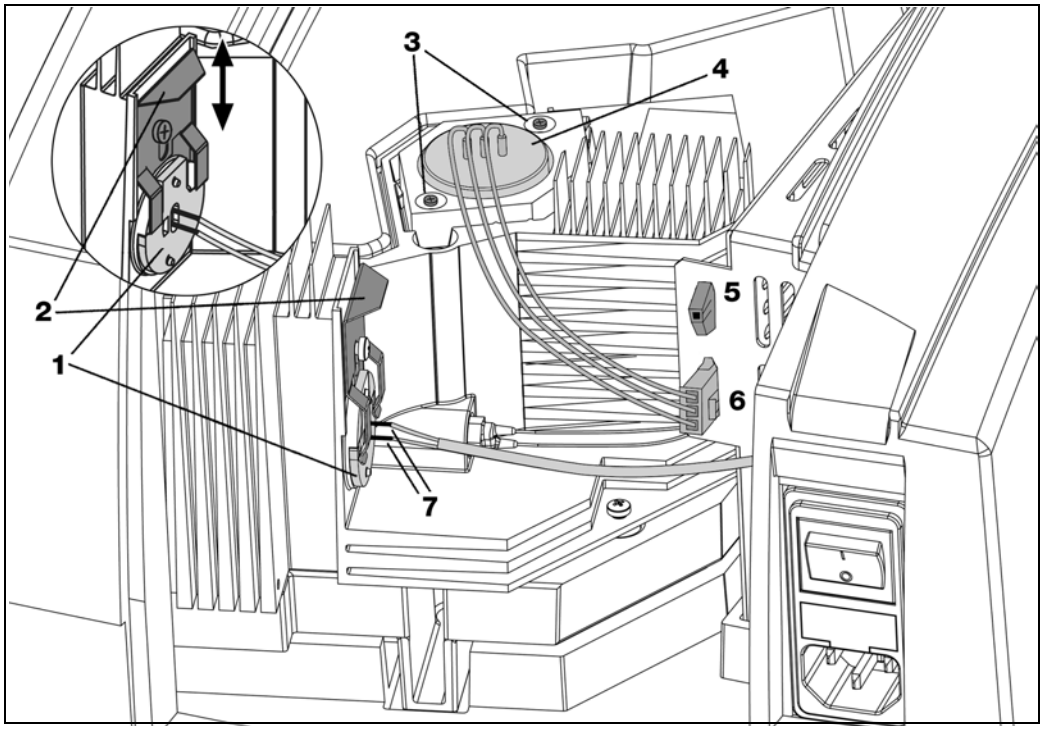
Other information

It is important that the Flowcell never be left to air dry. The Flowcell should be flushed and filled with deionized water when not in use. The tubing should remain attached. When use is resumed, the Flowcell need only be emptied, flushed several times, the filled with sample.

At the end of the day, follow the daily cleaning procedure, described in "Cleaning Procedure - Daily" on page 140. Short cuts in the cleaning procedure usually prove to be self-defeating and should be avoided since a build up of contaminants may occur.

15.2 Changing Lamps

Figure 15.1 Lamp Compartment



1	Tungsten lamp
2	Spring
3	Screws
4	Deuterium lamp (lamp socket)
5	Plug contacts for fan
6	Plug contact for Deuterium Lamp
7	Contacts of tungsten lamp with cable

Changing the Tungsten Lamp

1. Switch the instrument off.
2. Unplug the power cord.

WARNING The instrument must remain switched off until you have finished changing the lamp.

WARNING Wait until the lamp has cooled.

3. Use a screwdriver to remove the cover from the back of the instrument (the screws may be slotted or cross-headed).
4. Place the cover and the attached fan carefully beside the instrument (take special care with the fan cable).

5. Push the spring (2) up and remove the tungsten lamp (1) (Cat. No: A23778) from the lamp compartment.

6. Unplug both plug contacts (6) from the tungsten lamp.

WARNING Hold the lamp only by the lamp socket. Avoid touching the glass!

7. Push these plug contacts firmly onto the new tungsten lamp.

8. Insert the tungsten lamp into the lamp compartment. Push the spring down.

9. Check that the spring and the lamp socket are positioned correctly.

10. Use a screwdriver to screw the back cover onto the instrument again.

11. Plug in power supply again. The Instrument is now ready for use. Switch the instrument on.

12. Reset the Lamp History (see "Lamp History" on page 157).

Changing the deuterium lamp (UV)

1. Switch the instrument off.

2. Unplug the power cord.

WARNING The instrument must remain switched off until you have finished changing the lamp.

WARNING Wait until the lamp has cooled.

3. Use a screwdriver to remove the cover from the back of the instrument (the screws may be slotted or cross-headed).

4. Place the cover and the attached fan carefully beside the instrument (take special care with the fan cable).

5. Unplug the deuterium lamp (4) (Cat. No: A23792) from the socket by pushing down on the safety contact.

6. Use a screwdriver to unscrew the two fastening screws (3) (the screws may be slotted or cross-headed) out of the socket.

7. Holding the lamp socket, lift the deuterium lamp up out of the lamp compartment (remove the complete unit, including the cable).

8. Carefully insert the new deuterium lamp into the lamp compartment.

WARNING Do not touch the glass envelope on the new lamp. If it is touched, clean with alcohol.

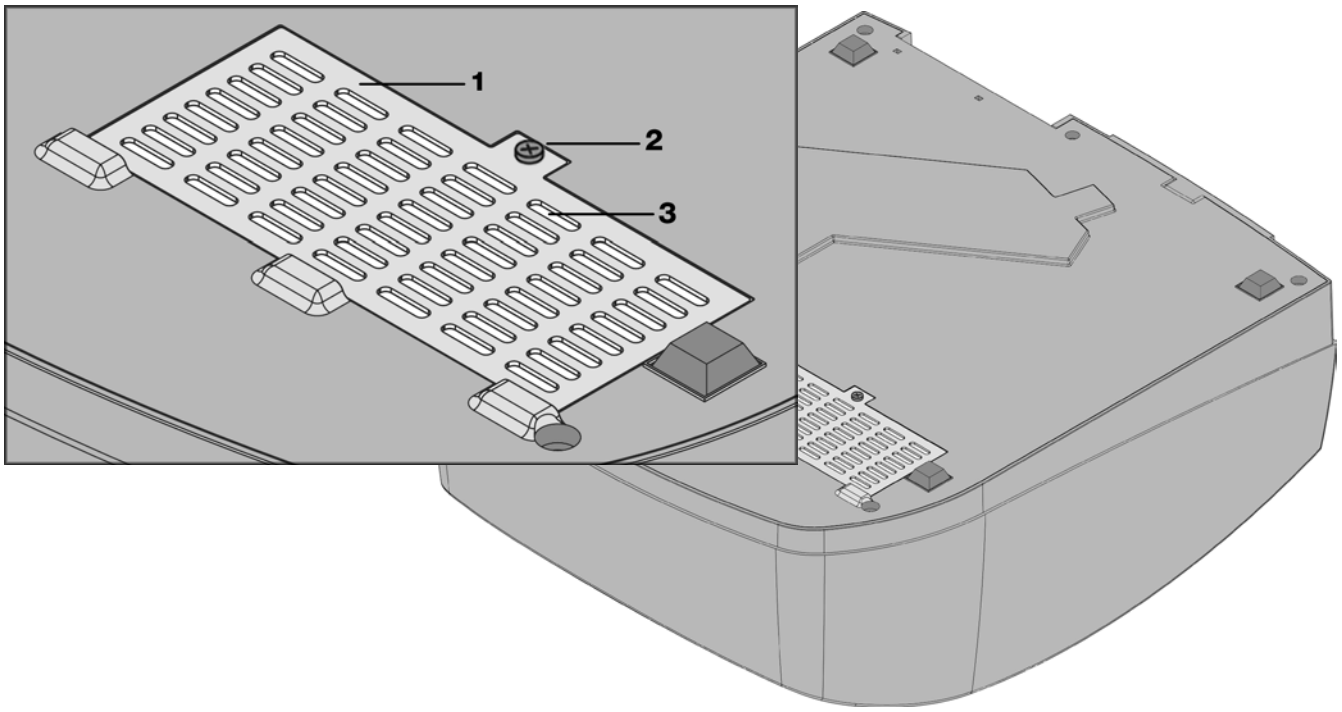
9. Screw in both fastening screws again until they are finger-tight.
10. Insert the deuterium lamp in the socket so that the safety contact clicks into place.
11. Use a screwdriver to screw the back cover onto the instrument again.
12. Plug in power supply again. The Instrument is now ready for use. Switch the instrument on.
13. Reset the Lamp History (see "Lamp History" on page 157).

15.3 Changing the Fuse

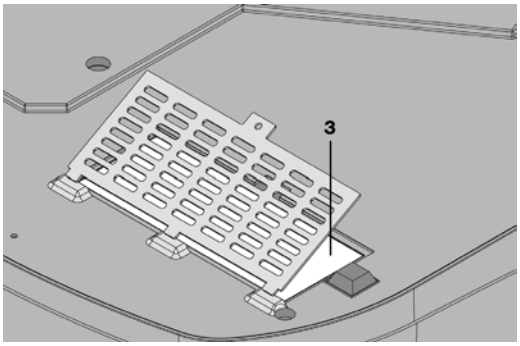
1. Switch the instrument off.
2. Unplug from the mains electricity supply.
3. Pull out the plastic base over the socket for power cable.
4. Remove the defective fuse (see Figure 2.3 on page 8).
5. Insert the new fuse (T 2 A L; 250 V).
6. Replace the plastic base.
7. Plug in to the main electricity supply.
8. Switch the instrument back on.

15.4 Changing the Filter Pad

Figure 15.2 Base of DU Series 700 Spectrophotometer with Filter Grid



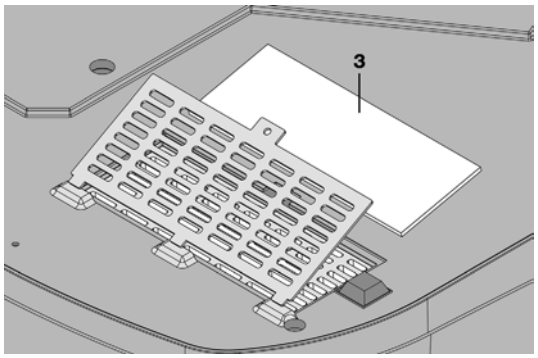
1	Filter grid
2	Phillips screw
3	Filter pad



NOTE Empty the cell compartment!

NOTE Unplug the instrument!

1. Turn the instrument over carefully and place it on a soft surface.
2. Use a screwdriver (standard or cross-head) to open the filter grid (1).
3. Lift the filter grid (1).



4. Remove the old filter pad (3) and replace it with a new one.
5. Screw the grid back in place.
6. Carefully stand the instrument upright.
7. Plug the instrument in. It is now ready for use again.

Troubleshooting

16.1 Power Up Diagnostics

Each time you power up the instrument, it performs a series of diagnostic tests automatically. These include:

- System Tests
- Memory Tests
- Voltage Tests
- λ–Calibration
- Lamp Tests
- Filter Adjustment

The Self Check display lists these tests. When each test completes successfully, the screen adds a check mark after the test name. If any of the tests do not finish successfully, the screen displays an error message.

When the screen displays an error message, take the recommended action that corresponds to the error message show in the following table (see "Error Messages" on page 148). If any of the tests fail and the screen does not display an error message, switch off the instrument, then switch it on again.

16.2 Error Messages

Table 16.1 Power-Up Error Messages

Error Message	Recommended Action
BEAM BLOCKED?	Remove whatever is blocking the beam in the sampling module and press the [START] soft key.
LAMP ALIGNMENT	Replace the visible lamp (see "Changing Lamps" on page 142).
LAMP OUT	Replace the visible lamp (see "Changing Lamps" on page 142).
LAMPS OVERHEATED	Not enough air is allowed to flow near the source compartment. Remove papers or other debris that are near the air vents on the back and underside of the instrument. Press the [START] soft key.
LID OPEN?	Close sampling module lid and press the [START] soft key.
MODULE REQUIRED	Insert sampling module and secure with module lock.
OFFSET CORRECTION	Close sampling module lid and press the [START] soft key.
UV LAMP OUT	Replace the deuterium lamp (see "Changing Lamps" on page 142). Clear the message by pressing the [CLEAR WARNING] soft key. It is possible to continue taking readings with the visible lamp, only.
VIS LAMP OUT	Replace the visible lamp (see "Changing Lamps" on page 142). Clear message by pressing the [CLEAR WARNING] soft key

16.3 Other Error Conditions

Table 16.2 Other Error Conditions

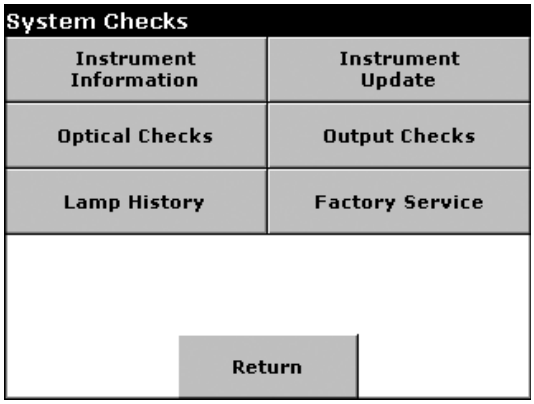
Error Message	Recommended Action
The instrument will not power up.	Verify that the power cord is securely plugged into the power outlet and the back of the instrument. Check fuses (see "Changing the Fuse" on page 144).
The screen is not readable.	Turn the contrast knob to the right of the screen until the screen is clear.
The wavelength is not accurate.	The instrument was powered up with a sample solution in the sampling module. Remove the sample and power up the instrument again.
Graphics are not printed properly.	The communications parameters are not set properly or graphics are not compatible with the selected printer.
Instrument locks up during printing.	Verify that the OUTPUT mode is turned off, if no device is available to accept data.

System Check

The DU Series 700 Spectrophotometer lets you verify instrument performance. The instrument contains the programs for checking wavelength accuracy, bandwidth, photometric noise, stray light, photometric accuracy, photometric repeatability, and photometric stability.

17.1 Call System Check from the Main Menu

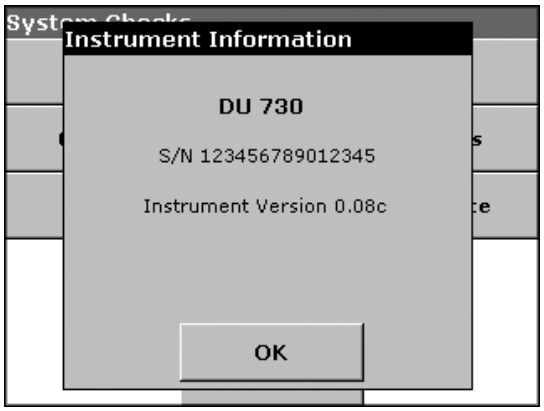
You must provide the samples necessary to perform the stray light and photometric accuracy and photometric repeatability tests.



Touch **System Checks** in the Main Menu to display the System Checks screen.

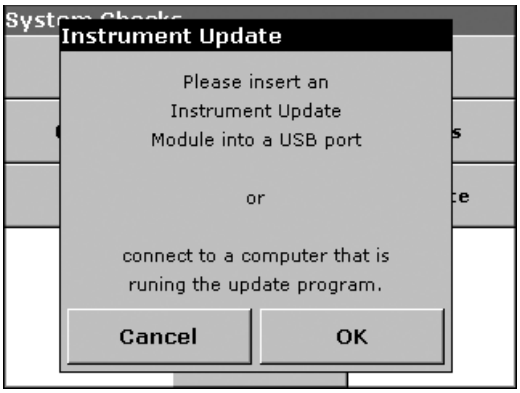
The System Checks screen allows you to get system information and run a number of performance tests.

17.2 Instrument Information



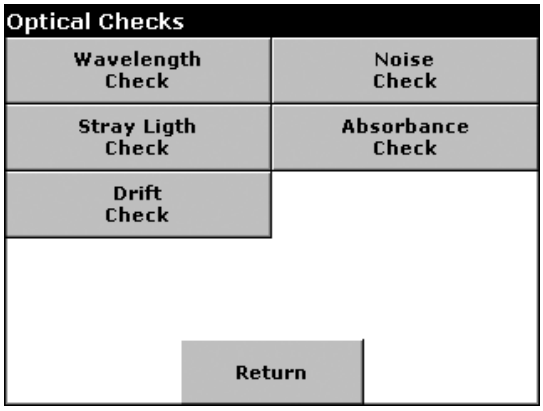
Touch **Instrument Information** to display the model, serial number, and software version.

17.3 Upgrading the Instrument Software



1. Touch **Instrument Update** to update the system firmware through the USB connection.
2. Follow the instructions on the screen and run the update program.
3. Touch **OK** to return to the System Checks screen.

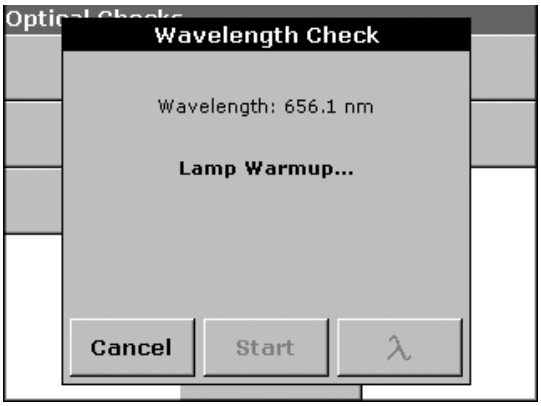
17.4 Optical Checks



Touch **Optical Checks** in the System Checks screen.

From the Optical Checks screen, you can execute a number of performance tests. These tests are described in the following sections.

Wavelength Check

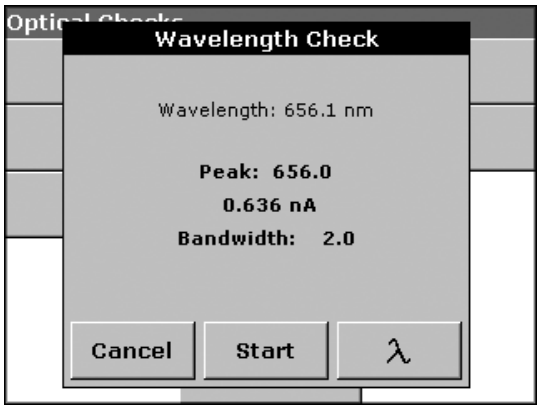


The Wavelength Check test lets you verify wavelength accuracy at 656.1 or 486.0 nm

When you select 656.1 nm, the test also reports the bandpass at 656.1 nm.

1. Touch λ to select the wavelength.
2. Make a choice and confirm with **OK**.
3. Touch **Start**.

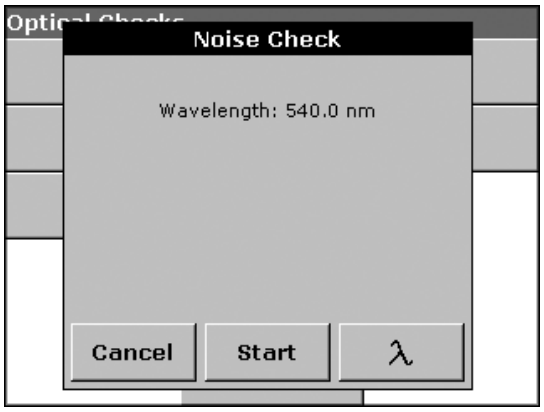
NOTE This might take awhile if the lamp is not warmed up sufficiently. In this case, the "Lamp Warmup..." message appears, informing you that the system is warming up the lamp to achieve the best results.

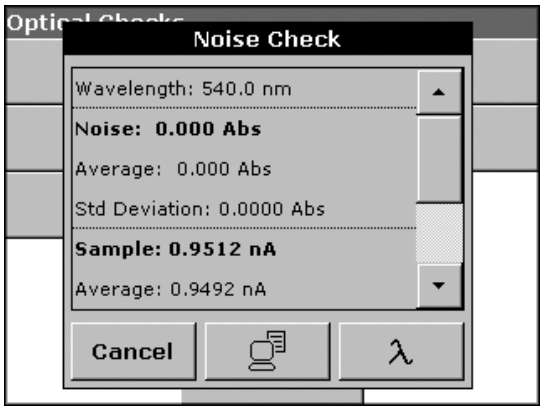


When complete, the instrument takes the readings and displays the results.

4. Touch **Cancel** to return to the Optical Checks screen.

Noise Check



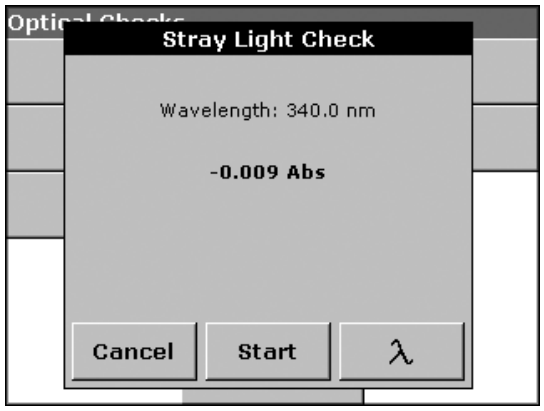


The Noise Check test lets you test the photometric noise in the instrument. The noise is specified at 546.1 nm at 0.0 A. However, you can also use this to test noise at any wavelength and at an absorbance level determined by a sample.

1. Touch λ to enter the wavelength.
2. Enter the wavelength using the numeric keypad and confirm with **OK**.
3. Touch **Blank**.
4. After the system has blanked, touch **Start**.

   When the test is complete, the screen displays the results. The instrument averages thirty readings for the blank and calculates the Average and Standard Deviation from 100 consecutive absorbance readings.
   - Noise
   - Sample
   - Reference
5. Touch **Cancel** to return to the Optical Checks screen.

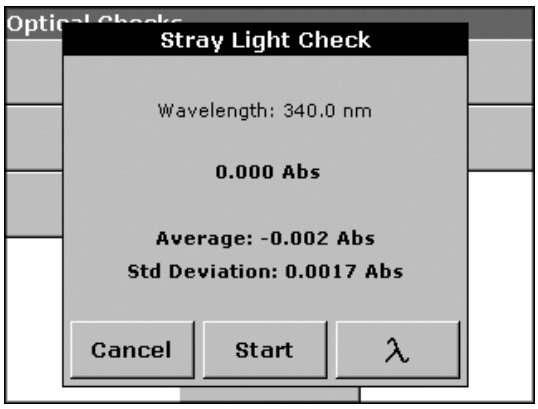
Stray Light Check



The Stray Light Check lets you measure the stray light in the instrument at 220 nm or 340.0 nm.

This test requires user-prepared samples. The test at 220.0 nm requires a 10 g/L Sodium Iodide (NaI) aqueous solution. The test at 340.0 nm requires a 50 g/L aqueous solution of Sodium Nitrite ($NaNO_2$). Both of these solutions need to be measured in quartz cuvettes, which transmit UV light.

1. Touch λ to select the wavelength.
2. Make a choice and confirm with **OK**.
3. Touch **Blank**.
4. After the system has blanked, insert the sample cuvette and touch **Start**.

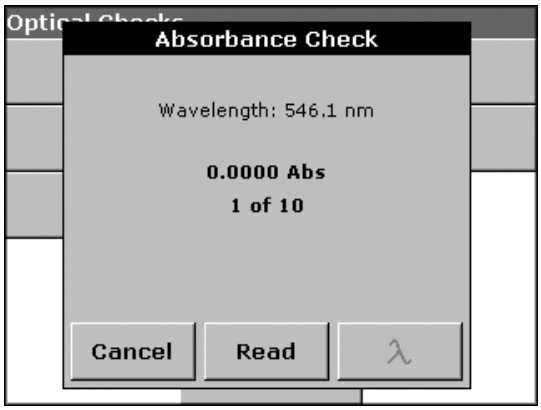


When the test is complete, the screen displays the results. The instrument averages thirty readings for the blank and calculates the Average and the Standard Deviation from 100 consecutive absorbance readings. The average of the readings is the stray light value in absorbance.

The stray light is typically about 3.3A, which is the same as 0.05%T.

5. Touch **Cancel** to return to the Optical Checks screen.

Absorbance Check

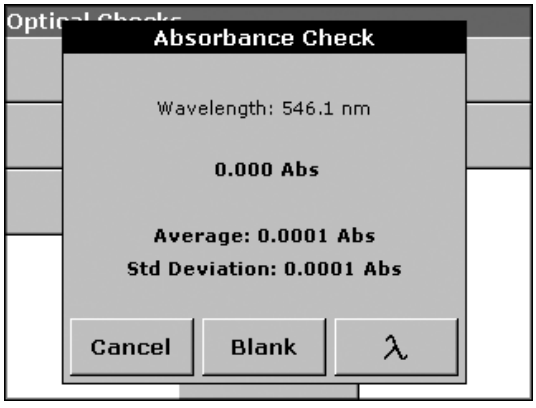


The Absorbance Check lets you test the photometric accuracy and repeatability of the instrument. This test requires standard filters, such as NIST SRM 2031 or 930e. The photometric accuracy is specified at 546.1 nm at 1.0A.

1. Touch λ to enter the wavelength.
2. Enter the wavelength using the numeric keypad and confirm with **OK**.

NOTE The system must read 10 replicates of a blank and readings from the sample solution to calculate the results.

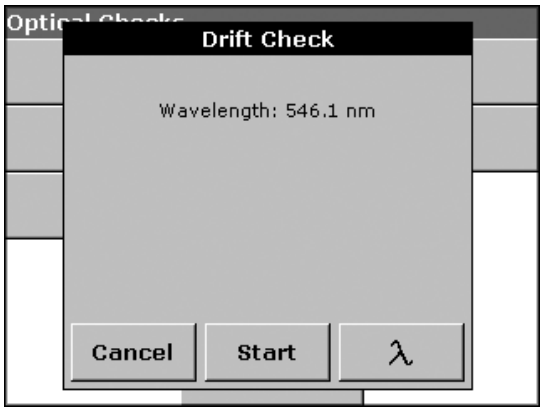
3. Insert the appropriate cuvette and touch **Blank** and then **Read**. This must be repeated 10 times.



The screen displays the results.

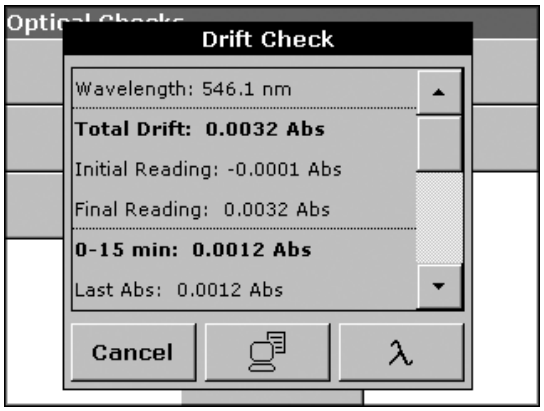
4. Touch **Cancel** to return to the Optical Checks screen.

Drift Check



The Drift Check lets you test the stability of the instrument.

1. Touch λ to enter the wavelength.
2. Enter the wavelength using the numeric keypad and confirm with **OK**.
3. Touch **Start**.

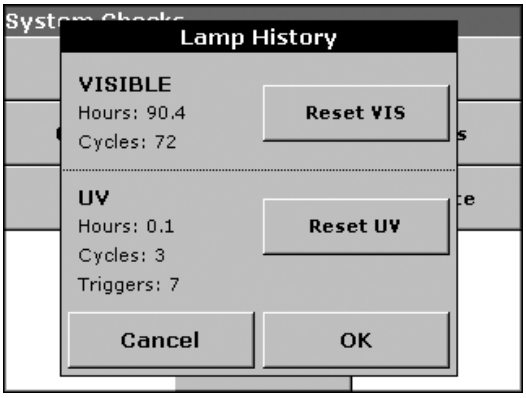


This test takes a reading every minute for one hour. Every fifteen minutes, the instrument uses linear regression to calculate the slope (rate of change) for the previous 15 minute interval.

The screen displays the last reading and slope for each 15 minute interval. At the end of the hour, the instrument calculates and displays the overall values.

4. Touch **Cancel** to return to the Optical Checks screen.

17.5 Lamp History



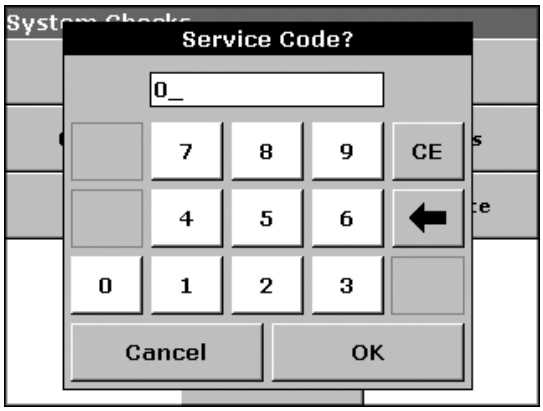
The Lamp History provides the following information:

- The amount of time that the lamp has been on (Hours).
- The number of times that the lamp has been turned on (Cycles).
- The number of times that the instrument tried to turn the UV lamp on (triggers).

After reviewing the lamp history:

- Touch **Reset VIS** to reset the Visible Lamp.
- Touch **Reset UV** to reset the UV Lamp.
- Touch **OK** to return to the System Checks screen.

17.6 Factory Service



The Factory Service requires a code.

NOTE These tests are not intended for customer use.

Specifications

NOTE Specifications are subject to change without notice.

18.1 Performance Specifications

Operating Mode:	Transmittance (%), Absorbance, and Concentration
Source Lamp:	Tungsten (visible) and Deuterium (UV)
Wavelength Range:	190 - 1100 nm
Wavelength Accuracy:	± 1 nm in Wavelength Range 200 - 900 nm
Wavelength Resolution:	Selectable (0.1, 0.2, 0.5, 1.0, 2.0, and 5.0 nm)
Wavelength Calibration:	Automatic
Wavelength Selection:	Automatic, based on method selection
Scanning Speed:	Depending on selected resolution (100-4500 nm/min)
Spectral Bandwidth:	3 nm
Photometric Readout:	-0.3 to 3.0 A or 0.1 to 100 %T
Photometric Accuracy:	± 0.005 A at 0.0 to 0.5 A 1% at 0.5 to 2.0 A
Photometric Linearity:	< 0.5% at 2.0 A (1% at > 2.0 A
Stray Light:	KI Solution at 220 nm > 3.3 Abs DU 700 is < or = 0.05 %T max at 220 and 320 nm

18.2 Physical and Environmental Specifications

Width:	17.7 in (450 mm)
Height:	7.9 in (200 mm)
Depth:	19.7 in (500 mm)
Weight:	34.2 lbs (15.5 kg)
Operating Requirements:	10 - 40°C (50 - 104°F), max. 90% relative humidity (non-condensing)
Storage Requirements:	-25 - 60°C (-13 - 140°F) max. 80% relative humidity (non-condensing)

18.3 Power and Other Specifications

Power:	100 - 120 V~; 200 - 240 V~; 50/60 Hz; automatic changeover
Ports:	USB 1.1
Cable:	USB cable < or = 3 meters **WARNING** To ensure compliance with EN 61326 and USB 1.1 requirements, the USB cable must not exceed 3 meters.
Water-resistant to	IP 32

Replacement Parts, Service Centers, and Warranty

19.1 Replacement Parts, Supplies, and Accessories

Table 19.1 DU Series 700 Instruments

A23615	DU-720 General Purpose UV/Vis Spectrophotometer
A23616	DU-730 Life Science UV/Vis Spectrophotometer

Table 19.2 Sampling Modules

A23619	Sipper Sampling Module
A23621	Peltier Temperature Control Module

Table 19.3 Cell Holders

A24211	Standard Cell Holder
A23620	Carousel with 7 Cell Positions
A23622	50 μL Microcell Holder
A23623	Turbidity Cell Holder
A23618	Multicell Holder

Table 19.4 Standard 1 cm Cuvettes

886506	Open Top, Optical Glass, 1 each
75152	Open Top, Optical Glass, 4 each
596492	Open Top, Optical Glass, 6 each
580012	Open Top, UV Silica, 1 each
596493	Open Top, UV Silica, 6 each
580015	Stoppered, UV Silica, 1 each
596494	Stoppered, UV Silica, 6 each

Table 19.5 Standard 1 cm Semi-Microcells, Masked

533041	2.0 mm Pathwidth, UV Silica, 2 each
533040	2.0 mm Pathwidth, UV Silica, 4 each
533043	4.0 mm Pathwidth, UV Silica, 2 each

Table 19.5 Standard 1 cm Semi-Microcells, Masked

533042	4.0 mm Pathwidth, UV Silica, 4 each
517056	4.0 mm Pathwidth, UV Silica, 6 each

Table 19.6 50 µL Microcells

523270	50 µL Microcell, 1 each
523450	50 µL Microcell, 2 each
523451	50 µL Microcell, 4 each
523452	50 µL Microcell, 6 each

Table 19.7 Long Pathlength 5 cm Cuvette

886510	Open Top, Optical Glass, 1 each

Table 19.8 Data Output Accessories

977510	HP DeskJet Print w/ USB Port and PCL3, 100-240VAC
512949	USB Printer Cable

Table 19.9 Replacement Parts and Supplies

A23778	Halogen Lamp Replacement Kit
A23792	Deuterium Lamp Replacement Kit
A23772	Fuse, Time Delay, 250V, 2.5A

Table 19.10 Replacement Parts for Sipper Sampling Module

A24209	750 µL Flow Cell
A23800	Flow Cell Tubing Kit, inlet and waste
A23801	Peristaltic Pump Tubing Kit, set of 4
586656	Waste Bottle

Table 19.11 Trace-Klean™ Solution

589784	Concentrated, 946 mL
598190	Dilute, 473 mL

Table 19.12 User's Guides

A24014	DU Series 700 User's Guide, English
A24015	DU Series 700 User's Guide, French
A24016	DU Series 700 User's Guide, Italian
A24017	DU Series 700 User's Guide, German
A24018	DU Series 700 User's Guide, Spanish
A24019	DU Series 700 User's Guide, Japanese

19.2 Repair Service/Beckman Coulter Sales and Service Offices

Technical support and service in the United States:

Beckman Coulter, Inc.
4300 N. Harbor Boulevard
P.O. Box 3100
Fullerton, CA 93834-3100
Tel:(800) 742-2345
Fax:(800) 643-4366

For up-to-date information on offices worldwide, please go to:

http://www.beckmancoulter.com/hr/contactus/default.asp

19.3 Warranty

Subject to the exceptions and upon the conditions stated below, Beckman Coulter warrants that this product shall be free from defects in workmanship and materials for one year after delivery of the products to the original Buyer by Beckman Coulter, and if any such product should prove to be defective within such one-year period, Beckman Coulter agrees, at its option, either (i) to correct by repair or, at Beckman Coulter's election, by replacement with equivalent product any such defective product, provided that investigation and factory inspection discloses that such defect developed under normal and proper use, or (ii) to refund the purchase price. The exceptions and conditions mentioned above are as follows:

(a) components or accessories manufactured by Beckman Coulter which by their nature are not intended to and will not function for ninety days are warranted only to give reasonable service for a reasonable time; what constitutes reasonable time and reasonable service shall be determined solely by Beckman Coulter. A complete list of such components and accessories is maintained at the factory.

(b) Beckman Coulter makes no warranty with respect to components or accessories not manufactured by it; in the event of defect in any such component or accessory Beckman Coulter

will give reasonable assistance to Buyer in obtaining from the respective manufacturer whatever adjustment is authorized by the manufacturer's own warranty.

(c) any product claimed to be defective must, if required by Beckman Coulter, be returned to the factory, transportation charges prepaid, and will be returned to Buyer with transportation charges collect unless the product is found to be defective, in which case Beckman Coulter will pay all transportation charges.

(d) Beckman Coulter shall be released from all obligations under all warranties, either expressed or implied, if any product covered hereby is repaired or modified by persons other than its own authorized service personnel unless such repair by others is made with the written consent of Beckman Coulter.

IT IS EXPRESSLY AGREED THAT THE ABOVE WARRANTY SHALL BE IN LIEU OF ALL WARRANTIES OF FITNESS AND OF THE WARRANTY OF MERCHANTABILITY AND THAT BECKMAN SHALL HAVE NO LIABILITY FOR SPECIAL OR CONSEQUENTIAL DAMAGES OF ANY KIND OR FROM ANY CAUSE WHATSOEVER ARISING OUT OF THE MANUFACTURE, USE, SALE, HANDLING, REPAIR, MAINTENANCE OR REPLACEMENT OF THIS PRODUCT.

Representations and warranties made by any person, including dealers and representatives of Beckman Coulter, which are inconsistent or in conflict with the terms of this warranty, shall not be binding upon Beckman Coulter unless reduced to writing and approved by an expressly authorized officer of Beckman Coulter.